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.

Biochemical Pharmacology 185 (2021) 114431

Contents lists available at ScienceDirect

Biochemical Pharmacology

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

Review

PDE4 inhibition as a therapeutic strategy for improvement of pulmonary dysfunctions in Covid-19 and cigarette smoking

Claire Lugnier [a,*], Hayder M. Al-Kuraishy [b], Eric Rousseau [c]

[a] Directeur de Recherche 1 CNRS/université de Strasbourg, Institut de Physiologie, Faculté de Médecine, CRBS, UR3072: "Mitochondrie, stress oxydant et protection musculaire", 1 rue Eugène Boeckel, 67000 Strasbourg, France
[b] Medical Faculty College of Medicine, Al-Mustansiriya University, P.O. Box 14132, Baghdad, Iraq
[c] Department of Obstetrics and Gynecology, Faculty of Medicine and Health Sciences, Université de Sherbrooke, and Centre de Recherche du CHUS, Sherbrooke, QC, Canada

ARTICLE INFO

Keywords:
COVID-19
Cigarette smoking (CS)
Angiotensin-converting enzyme 2 (ACE2)
PDE4
Inflammation
Respiratory epithelial cells

ABSTRACT

Angiotensin-converting enzyme 2 (ACE2) is the binding-site and entry-point for SARS-CoV-2 in human and highly expressed in the lung. Cigarette smoking (CS) is the leading cause of pulmonary and cardiovascular diseases. Chronic CS leads to upregulation of bronchial ACE2 inducing a high vulnerability in COVID-19 smoker patients. Interestingly, CS-induced dysregulation of pulmonary renin-angiotensin system (RAS) in part contributing into the potential pathogenesis COVID-19 pneumonia and acute respiratory distress syndrome (ARDS). Since, CS-mediated ACE2 activations is not the main pathway for increasing the risk of COVID-19, it appeared that AngII/AT_1R might induce an inflammatory-burst in COVID-19 response by up-regulating cyclic nucleotide phosphodiesterase type 4 (PDE4), which hydrolyses specifically the second intracellular messenger 3′, 5′-cyclic AMP (cAMP). It must be pointed out that CS might induce PDE4 up-regulation similarly to the COVID-19 inflammation, and therefore could potentiate COVID-19 inflammation opening the potential therapeutic effects of PDE4 inhibitor in both COVID-19-inflammation and CS.

1. Introduction

The severe acute respiratory syndrome (SARS) started in China 2003, caused by a specific type of coronavirus named SARS-CoV-2, that might be related to emerging infectious diseases leading to economic and health global burden. Later on, Middle East respiratory syndrome (MERS) caused by MERS-CoV was emerged in 2012 [1]. In 2019, a pulmonary infection caused by a novel coronavirus nCoV-19 also called SARS-CoV-2 was started in Wuhan, China led to coronavirus infection disease (COVID-19), which was initially named Wuhan pneumonia, that seems to be originating from bats [2].

The binding site and the entry-point for SARS-CoV-2 is angiotensin-converting enzyme 2 (ACE2), which is highly expressed in the respiratory epithelial cells, lymphocytes, endothelial cell, and renal tubular epithelial cells. The clinical presentations of COVID-19 are mild in 81%, asymptomatic in 50% and 2–4% severe likes acute respiratory distress syndrome (ARDS), acute kidney injury, sepsis and coagulopathy [3]. According to Shurin et al., the immune responses induced by SARS-CoV-2 infection seem to be in two-stages. As most of the infected individuals develop only mild or no clinical symptoms, it is conceivable that during the incubation and non-severe stages, a specific adaptive immune response is required to eliminate the virus and to preclude disease progression to severe stages [4]. The case-fatality rate of COVID-19 patients is higher in patients with underlying comorbidities, including: hypertension, diabetes mellitus and tobacco smoking, which accelerate the viral entry or replication of SARS-CoV-2 [5].

The nicotine smoking, either from cigarette smoking (CS) or from electronic cigarette, is the leading cause of pulmonary and cardiovascular diseases. As well, CS leads to pulmonary hypertension, endothelial dysfunction, congestive heart failure, chronic pulmonary disease, and metabolic complications directly or through dis-regulation of the renin-angiotensin system (RAS) [6]. In another way, it is well known that intracellular signaling, mediated by cyclic nucleotide phosphodiesterases (PDEs, PDE1-PDE11), controls tissue cAMP and cGMP levels in response to receptor activation. Interestingly, it is largely implicated in lung and cardiovascular diseases as well as in inflammation, notably ARDS [7–10]. Therefore, RAS and PDEs together might open new therapeutic approaches in COVID-19, notably in CS.

* Corresponding author.
E-mail address: claire.lugnier@unistra.fr (C. Lugnier).

https://doi.org/10.1016/j.bcp.2021.114431
Received 23 November 2020; Received in revised form 15 January 2021; Accepted 19 January 2021
Available online 28 January 2021

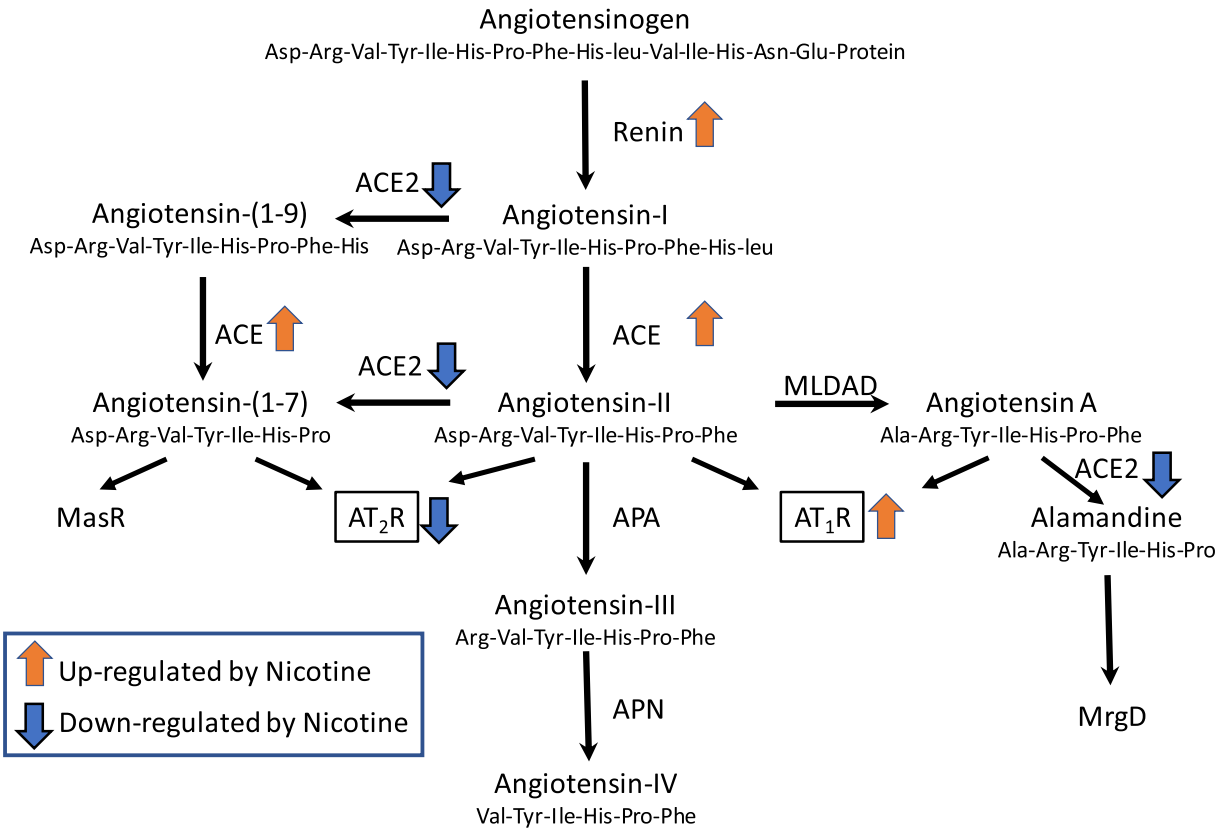

Fig. 1. Effect of nicotine smoking on the renin-angiotensin system (RAS). Angiotensinogen is hydrolyzed by renin to produce angiotensin (ANG) I, which is then converted by angiotensin-converting enzyme (ACE) into the biologically active ANG II. More recently, renin- and ACE-independent formation of ANG II have also been reported. By cleaving ANG II into ANG-(1–7), ACE2 plays a pivotal role in the compensatory ACE2/ ANG-(1–7)/MasR axis of the RAS by counterbalancing the deleterious actions of the ACE/ANG II/AT_1R arm. ACE: Angiotensin converting enzyme; ACE2: Angiotensin converting enzyme type 2; MLDAD: mononuclear leukocyte-derived aspartate decarboxylase; MasR: Mass receptor; AT_1R: Angiotensin II type 1 receptor; AT_2R: Angiotensin II type 2 receptor; APA: Aminopeptidase A; APN: Aminopeptidase N; MrgD: Mas-related G protein-coupled receptor D. Adapted from[11].

2. The Renin-Angiotensin system (RAS)

RAS is composed of different peptides involved in the regulation of blood pressure and homeostasis. The hepatic angiotensinogen is hydrolyzed by renin to angiotensin-I (Ang-I), which is converted by the angiotensin converting enzyme (ACE) to angiotensin-II (Ang-II) that activates angiotensin-II type 1 receptor (AT_1R). Ang-II is further hydrolyzed to Ang-III and Ang-IV by aminopeptidase A (APA) and aminopeptidase N (APN), respectively. ACE2 hydrolyzed Ang-I to Ang_{1-9} and Ang-II to Ang_{1-7} which are potent vasodilators *via* activation of angiotensin-II type 2 receptor (AT_2R), and mass receptor (MasR) (Fig. 1) [11]. Indeed, both nicotinic receptor and ACE2 are expressed in the pulmonary alveolar type II cells, epithelial cells, and pulmonary macrophages. Besides, pulmonary micro-vascular endothelial cells have a higher expression of ACE, which participates in the regulation of systemic blood pressure [12]. AT_1R and AT_2R are broadly distributed in the lung. AT_1R is mainly localized at stromal fibroblasts, macrophages and vascular smooth muscle cells, while, AT_2R is chiefly found in brush borders of bronchial epithelium in addition to pulmonary macrophages and endothelial cells [13]. It has been reported by different studies that pulmonary RAS is linked in the pathogenesis of different pulmonary disorders unrelated to hypertension or body fluid overloads, such as ARDS, acute lung injury (ALI), pulmonary inflammation and fibrosis [14].

3. Renin-Angiotensin system (RAS) and cigarette smoking (CS)

The lung is the first organ which encounters the nicotine. Of interest, Glynos et al. reports that both e-cigarette vaping and conventional CS negatively impact lung biology, triggering inflammatory responses and adversely respiratory system mechanisms [15]. Furthermore, it has been reported that in SARS-CoV-2, that e-cigarette induces pulmonary inflammation and dis-regulated repair and increased SARS-Cov-2 Covid-19 ACE2 receptor (ACE2)[16]. Although, nicotine per se might be beneficial in COVID-19, by inhibiting nicotinic acetylcholine receptor [17] or by decreasing ACE2 expression [18], CS affects pulmonary RAS in different ways. In one way, Lu et al., found that CS increases the expression of ACE and elevates the circulating Ang-II concentrations, which is not sustained following ten days of the effect. However, in healthy human, ACE activity increased for minutes only and returns to the normal after first exposure. As well, both nicotine and its metabolites elevate the *in vitro* activity of ACE in the cultured endothelial cells [19]. Of interest, it must be pointed out that such consumptions might induce intracellular signaling changes, possibly enzyme expression such as PDE4 expressions [20]. In another way, nicotine, related to CS, reduces the expression of ACE2 in the pulmonary smooth muscle cells, directly or through elevation of Ang-II, contributing to the initiation of pulmonary hypertension. Since, high Ang-II downregulates ACE2 through an AT_1R signaling pathway, therefore, Ang-II-receptor blockers might restore the activity of ACE2 of chronic smoker patients [11]. However, chronic CS leads to alveolar septal fibrosis and apoptosis through induction of oxidative stress *via* AT_1R-dependent pathway. This CS induced-pulmonary dysfunction might be attenuated by the administration of Ang_{1-7}, which acts on MasR [21] (see Fig. 1). Therefore, acute CS adversely affects the pulmonary RAS through up-regulation of ACE and AT_1R which might be accompanied by down-regulation of ACE2 and AT_2R. Podowski et al. showed that in mice losartan may protect lung tissue from cigarette smoke-induced oxidative stress levels and

normalize AT_1R expression in the lung parenchyma, pointing out the beneficial effect of losartan (3–30 mg/kg in mice for 2 months) in oxidative stress [22].

Regarding CS subjects and susceptibility to COVID-19, Berlin et al. found that pulmonary ACE2 gene expression was higher in the former smoker subjects, that may explain the higher vulnerability of smoker patients in the early reported cases of COVID-19 in China [23]. Besides epidemiological studies, it was observed that CS subjects are at a higher risk for SARS-CoV-2 infection and required mechanical ventilation and aggressive interventions [24]. Bai et al. reported that the affected COVID-19 patients were chiefly smokers, 21.2% compared with non-smokers, 14.5%, suggesting that CS is involved in the pathogenesis of COVID-19 due to suppressing of the pulmonary ACE2/Ang_{1-7}/MasR axis [25]. It was suggested that individuals 'nicotine primed' to have a higher risk because nicotine can directly impact the putative receptor for the virus (ACE2) and lead to deleterious signaling in lung epithelial cells [26]. Therefore, CS induced-deregulations of pulmonary RAS may in part contribute into the potential pathogenesis COVID-19 pneumonia and ARDS. CS induced up-regulation of pulmonary ACE2 is not regarded as the main pathway since, ACE2 activators, like xanthenone and diminazene, seem to be protective against ALI and COVID-19 pneumonia [27]. Besides the CS induced down-regulation of ACE2, the induction of oxidative stress is involved by ALI [28]. The underlying mechanism of CS induced over-expression of ACE2 is not through direct interaction, but nicotine activates pulmonary epithelia nicotinic receptors, which are co-expressed with pulmonary ACE2. Activation of nicotinic receptors leads to stimulation of signaling pathways, including TNF-α and p38/mitogen activated protein kinase (p38/MAPK) which induce protease activation, inflammatory signaling and apoptosis, which together disturbing pulmonary alveolar cell function with subsequent ACE2 activation [29]. Furthermore, a recent study clearly suggests that smoking may promote cellular uptake mechanisms of SARS-CoV-2 through α7-nAChR signaling with a downstream induction of phospho-AKT and phospho-p42/p44 MAPK [30].

These changes provoke ACE2 expression that has abnormal functions with higher binding affinity to SARS-CoV-2. These findings might explain the higher incidence of COVID-19 in chronic CS [31]. Cui et al. suggest that ACE2 inhibitors like azathioprine, valproic acid, butyrate, sambucus and epoxomicin are effective anti-SARS-CoV-2 through inhibition of its entry *via* ACE2 [32]. This study concludes that CS-mediated ACE2 activations is not the main pathway for increasing the risk of COVID-19, as ARBs and ACEIs also increase the expression of ACE2 without increasing the risk of COVID-19. Therefore, extensive researches are recommended to explore and clarify the links between CS and RAS in CS at molecular and sub-molecular levels. As much as, a meta-analysis reports that COPD patients with confirmed COVID-19 show a higher mortality rate at 60%, moreover a higher mortality rate of 38.5% was reported in current smokers with confirmed COVID-19 [33]. Moreover, a recent meta-analysis on 11,189 patients reports that smoking doubles the risk of COVID-19, although, this result being non-significative is probably related to different mechanisms, notably acting on immune system and COPD [34]. In that way, lastly and interestingly, a recent review reports for CS that current smokers show a reduced risk of SARS-CoV-2 infection, while former smokers appear to be at increased risk of hospitalization, increased disease severity and mortality from COVID-19 [35]. This might open a new therapeutic approach requiring to differently analyze CS clinical studies.

Nevertheless, beyond AT_1R receptor signaling, it must be of interest of taking into accounts the rapid cyclic nucleotides degradations, governed by PDEs, that play a major role in normal and pathophysiological intracellular signaling, notably in inflammation and oxidative stress, for which new potent and original PDE inhibitors have been developed [9,10].

Table 1
Effects of PDE4 inhibitors (IC_{50}, μM) on native PDE1 to PDE5.

Enzyme	PDE1	PDE2	PDE3	PDE4	PDE5
Substrate	cGMP/ Ca^{2+}	cAMP/ EGTA	cAMP/ EGTA	cAMP/ EGTA	cGMP/ EGTA
Modulator	CaM	+5μM cGMP			
Rolipram [77]	N.S.	N.S.	N.S.	1.2	N.S.
RP 73401 [45]	N.S.	N.S.	N.S.	0.001	N.S.
Roflumilast [87]	N.S.	N.S.	N.S.	0.0008	N.S.
Pentoxifylline [77]	N.S.	119	84	135	74

IC_{50} determined at 1 μM substrate concentration, N.S.: $IC_{50} > 200$ μM. CaM: calmodulin.

4. The PDE4 subfamily and chronic obstructive pulmonary diseases (COPD)

Since SARS mainly affects pulmonary system, which is the target of chronic obstructive pulmonary diseases (COPD), for which many new compounds have been conceived and some of which are successfully marketed as PDE inhibitors [7,9]. Thus, among the eleven-known PDE families, the PDE4 family, hydrolyzes specifically the cyclic 3′, 5′-AMP (cAMP) in 5′-AMP and H^+. PDE4 was characterized by its specific inhibitor rolipram [36], and was designed as a COPD therapeutic target. This family is pointed out by its great number of subfamilies (A, B, C and D), by its great number of variants (>25 human variants), being distributed in various tissues and subcellular compartments, allowing a fine tuning of compartmentalized intracellular signaling [7]. This family is particularly intricate with intracellular signaling components, including a great variety of kinases as well as peptides, altogether designed signalosome [7,9,10]. The PDE4 family, early mainly implicated in inflammation, and oxidative stress was recognized as a new target for COPD treatments, for which pharmaceutical industry developed third generations of new very specific and potent PDE4 inhibitors, such as roflumilast with low emetic effects [9]. PDE4, as RAS, is largely present in the lung (airway epithelium [37]; airway smooth muscle [38]; pulmonary vessels [39]; endothelium [40,41]; and inflammatory cells [42]. Therefore, PDE4 inhibitors are prone for the treatment of inflammatory diseases [42]. In that context, a third's generation compound, such as NCS 613, was designed and synthesized as a specific PDE4 inhibitor ($IC_{50} = 42$ nM) [43], (Table 1). Interestingly, this compound did not induce emesis in rat (without and with pentagastrin) when administrated to until 30 mg/kg i.v. whereas, RP 73,401 (a second's generation compound from Rhône-Poulenc), induced emesis at 30 mg/kg in the absence of pentagastrin, and at 0.3 mg/kg in the presence of pentagastrin [44]. Altogether, the new PDE4 inhibitor, NCS 613, might be promising as a therapeutic compound in lung inflammation. In that way, NCS 613, inhibits *in vivo* lupus progression in MRL/lpr mice and inhibits *ex vivo* basal and LPS-induced TNF- α secretion by peripheral blood lymphocytes from MRL/lpr mice as well as from SLE patients, attesting the *in vivo* therapeutic potential of the NCS 613 [45]. Thus, Yougbare et al., reported that in human lung high cAMP-PDE activities were found in the cytosolic fractions from lung parenchyma and distal bronchi. Interestingly, these cAMP-PDE activities were related to PDE4 (rolipram sensitive) activity contributions by 40% and 56%, respectively. Bronchial PDE4 activity was 3.5-fold higher than its activity in pulmonary arteries, indicating its important implication in human pulmonary function. PDE4A, PDE4B, PDE4C, and PDE4D isoforms were differently detected by Western blots in all three subcellular fractions (cytosolic, microsomal, and nuclear). Moreover, immunostaining studies clearly showed that PDE4B and PDE4C isoforms were consistently detected by immunostaining in epithelial and arterial smooth muscle cells of bronchial cryo-sections [46]. Therefore, the effects of the new PDE4 inhibitor, NCS 613, were studied on human pulmonary tissues. In that way, the performed studies revealed that NCS 613 abolishes inflammation in

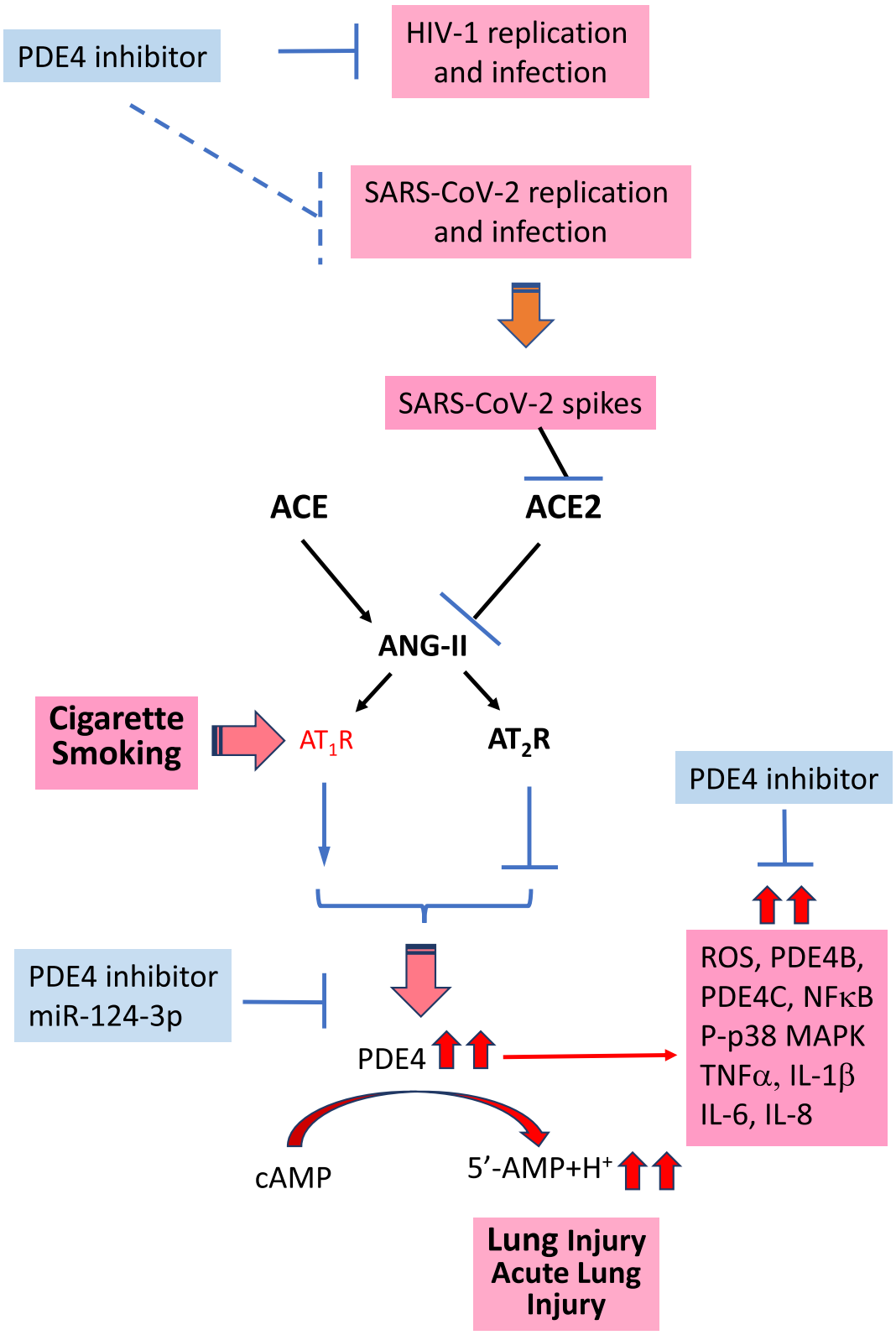

Fig. 2. Cross-talk between COV-19 and PDE4 mediated by AT_1R and its potentiation by CS. On one hand, PDE4 inhibitor overcomes HIV-1 infection and infection, and might also inhibit SARS-COV-2 replication and infection. On the other hand, SARS-COV-2 spikes by inhibiting ACE2 regulates ANG-II production, which induces PDE4 up-regulation. Cigarette smoking might up-regulate AT_1R and therefore increases PDE4. The up-regulated PDE4 might produce an increase of 5′-AMP, as well as of H^+, inducing lung injury and acute lung inflammation. These de-regulations might be related to PDE4B and PDE4C up-regulations, inducing increases in ROS production, as well in TNF-α, IL-1 β, IL-6, IL-8, NFκB and p-p38 MAPK. Interestingly, PDE4 is also up-regulated by CS, opening a commune therapeutic approach in COVID-19 and CS. Altogether, the use of specific PDE4 inhibitor or microRNA-124-3p to act on SARS-COV-2 and on PDE4 up-regulation, represents innovative approaches for treating Covid-19 CS.

the lung parenchyma by restoring IκBκ detection, reporting a potential buffering of NF-κB signaling. Moreover, this PDE4 inhibitor reduces human distal bronchial hyper-responsiveness by decreasing p-CPI-17, which in turn may be related to a down-regulation of PDE4C, PDE4B, and other intracellular signaling pathways such as p-p38-MAPK [46]. Furthermore, NCS 613 displays anti-inflammatory and anti-proliferative properties on A549 human lung epithelial cells and human lung adenocarcinoma explants [47], and suppresses systemic inflammation and immune complex deposition by increasing cAMP level [48]. These studies, clearly demonstrate the importance of PDE4 participation in the control of human lung responsiveness, inflammation and hyper-responsiveness processes, as well as of the targeted effectiveness of PDE4 inhibitors. Furthermore, NCS 613 treatment is also clearly effective on peripheral blood mononuclear cells (PBMCs) from both healthy donors and lupus patients. This specific PDE4 inhibitor decreased PDE4B whether it up-regulated PDE4C in human PBMCs which mainly control tissue inflammation. NCS 613 decreased p-p38 MAPK as well as NF-kB translocation to the nucleus and abolishes lipopolysaccharide (LPS)-induced inflammation by reducing IL-1β, IL-6, IL-8, and TNF- α cytokines [49]. In accordance, Chen et al., reported that the plasma cytokine levels of IL-6 and TNF- α were significantly increased in severe cases of patients with COVID-19, in comparison with moderate cases [50]. Altogether, these data obtained from human tissues and moreover inflammatory cells, show that PDE4s in lung and inflammatory cells might contribute in pulmonary disease, notably in SARS/ARDS.

It must be pointed out that losartan, used as an AT_1R antagonist in the RAS, inhibits the PDE4 activity ($IC_{50} = 26$ μM [51]; used at 10 μM to antagonize AT_1R [52]). Interestingly, a study reported that Ang-II, AT_1R agonist, induces PDE4 up-regulation and increases by 44% the PDE4A protein expression and mediates inflammation cascade and oxidative stress, opening a new way of interacting with RAS [53]. Altogether, these data clearly show that PDE4 might be an interesting target in COVID-19 inflammation and oxidative stress, as much as PDE4 is mainly present in pulmonary tissues inflammatory cells and endothelial cells [10]. Of interest, it was originally reported that miR-124-3p, which helps to protect against ARDS attenuate the increases in IL-1β, IL-6, and the TNF-α levels induced by LPS, suppressed LPS-induced p65 expression, an essential subunit of NF-κB, and thereby inhibit the inflammatory response in lung tissue and promotes the apoptosis of macrophages in a mouse model of ARDS [54]. Later, it was also reported that miR-124-3p attenuates severe community acquired pneumonia progression in macrophages by targeting TNF receptor-associated factor 6 [55]. In accordance with these anti-inflammatory effects, it was previously reported that miR-124-3p suppresses the activity of mTOR signaling, inhibits neuro-inflammation, mainly through overcoming the expression of PDE4B [56]. Furthermore, another study demonstrated that LPS increased levels of TNF- α, IL-1 β, enhanced NF-kB activity and p-p38 MAPK, which were attenuated by miR-124-3p over-expression, suggesting that miR-124-3p may serve as a therapeutic target for severe community-acquired pneumonia [57]. Altogether, these data indicate that miR-124-3p inhibits PDE4 expression which participates in ARDS inflammation, representing a novel therapeutic approach at the RNA level targeting PDE4 for treating ARDS.

Chronic CS induced-pulmonary dysfunction is attenuated by the administration of $Ang_{1\text{-}7}$, which acts on MasR. Therefore, acute CS adversely affects the pulmonary RAS through up-regulation of ACE and with down-regulation of ACE2 and AT_2R (see Fig. 1), whether, as shown previously, AT_1R up-regulation induces PDE4 up-regulation and consequently inflammatory processes [53]. However, chronic CS leads to up-regulation of the bronchial ACE2 as a compensatory mechanism which returns to the normal following quitting of smoking [58]. Nevertheless, it was reported for smokers with COPD that PDE4A4 is up-regulated in lung macrophages, and that concomitantly in PBMCs, PDE4B2 and PDE4A4 are also up-regulated, indicating that PDE4 inhibitors might be beneficial to smokers [59]. This was attested by Zuo et al. by various studies showing that CS reduces cAMP by activating PDE4 in cultured human bronchial epithelial cells [60] (Fig. 2).

5. PDE4 and viral infection

PDE4, not only participates in human intracellular signaling, but also might participate in viral infection. Since, it was previously shown that TNF-α enhances human immunodeficiency virus (HIV)-1 replication *in vitro,* the PDE4 inhibitor, rolipram was canonically shown to inhibit HIV-1 replication, and to decrease HIV-1 p24 antigen production in acutely HIV-1 infected PBMCs [61]. Thereafter, this rolipram inhibitory effect on HIV-1 replication was confirmed and extended to TNF-α production, NF-κB and NFAT activation, induced by T-cell activation in Jurkat and primary T cells [62]. Thus, a pivotal role of PDE4 in HIV-1replication was reported by Gallo and colleagues, showing that rolipram potently inhibited HIV-1 replication in cultures stimulated by anti-CD3 / CD28 ± Tat. Furthermore, this PDE4 inhibitor also abrogates the Tat-mediated increase in IL-2 production and CD41 T cell proliferation, attesting that rolipram overcomes both viral replication and inflammation [63]. Interestingly, Beavo and colleagues, showed that infection of $CD4^+$ memory T cells by HIV-1 requires the expression of PDE4, and that rolipram abolishes HIV-1 DNA nuclear import in memory T cells, pointing out the important contribution of PDE4 in viral infection [64]. Thus, it was reported that cAMP produced in Tregs is involved in the suppression of HIV-1gene activation and expression *in vivo* in humanized mice [65]. Altogether, theses data clearly show that PDE4 is implicated virus replication and T cell activations, designing PDE4 as a possible target in viral infection. Interestingly, an *in-silico* study allowed to conceive the compound **3i** that inhibits TNF- α *in vitro* ($IC_{50} = 5.14$ μM) and which binds with the SARS-CoV-2 N-terminal RNA binding domain residues [66]. Altogether, this opens a promising approach to treat simultaneously the COVID-19 by targeting both SARS-CoV-2 and inflammation related to PDE4.

6. Covid-19, RAS-CS and PDE4

As stated before, during 2013 a new coronavirus issued in China. Thus, it induced ARDS by using their spikes to target pulmonary system. Interestingly, previously in 2005, a paper issued in Nature Medicine demonstrated that angiotensin converting enzyme 2 (ACE2) plays a strategic role in SARS-CoV-2 induced lung injury [67]. This study clearly showed that ACE2 is a crucial SARS-CoV-2 receptor *in vivo*, and that both SARS-CoV-2 infection and the spike protein of the SARS-CoV-2 reduce ACE2 expression. Therefore, the injection of SARS-CoV-2 spikes into mice worsens acute lung failure *in vivo* which can be attenuated by blocking the renin-angiotensin pathway (see RAS paragraph, pages 3–5). Thus, it was done by using the AT_1R antagonist losartan, being a PDE4 inhibitor, which, as expected, attenuated acute severe lung injury and pulmonary edema in spike-Fc–treated mice. These results provide a molecular explanation why SARS-CoV-2 infections cause severe and often lethal lung failure. They might be extended to COVID-19, to open new therapeutic approaches, by targeting PDE4 which greatly increases in both viral infection [61] and severe lung injury [42] (Fig. 2).

Since, CS-mediated ACE2 activation is not the main pathway for increasing the risk of COVID-19 in CS, beyond the use of Ang-II-receptor blockers, which results are not clearly established, it would be of interest to take into accounts the pathological implications of PDE4 at viral and host levels and therefore to investigate the beneficial effects of PDE4 inhibitors in COVID-19 treatment.

Therefore, as stated before, it is clearly established that the PDE4 inhibitor rolipram inhibits HIV-1 replication, which is dependent on PDE4 expression, attesting the crucial role of PDE4 in viral infection. Interestingly, the possible PDE4 inhibitor effect on SARS-CoV-2 infection was addressed in 2021 by conceiving effective compounds, such as compound 3i, targeting both SARS-CoV-2 and inflammation related to PDE4 [66].

Nevertheless, the SARS-CoV-2 infection processes targeting ACE2,

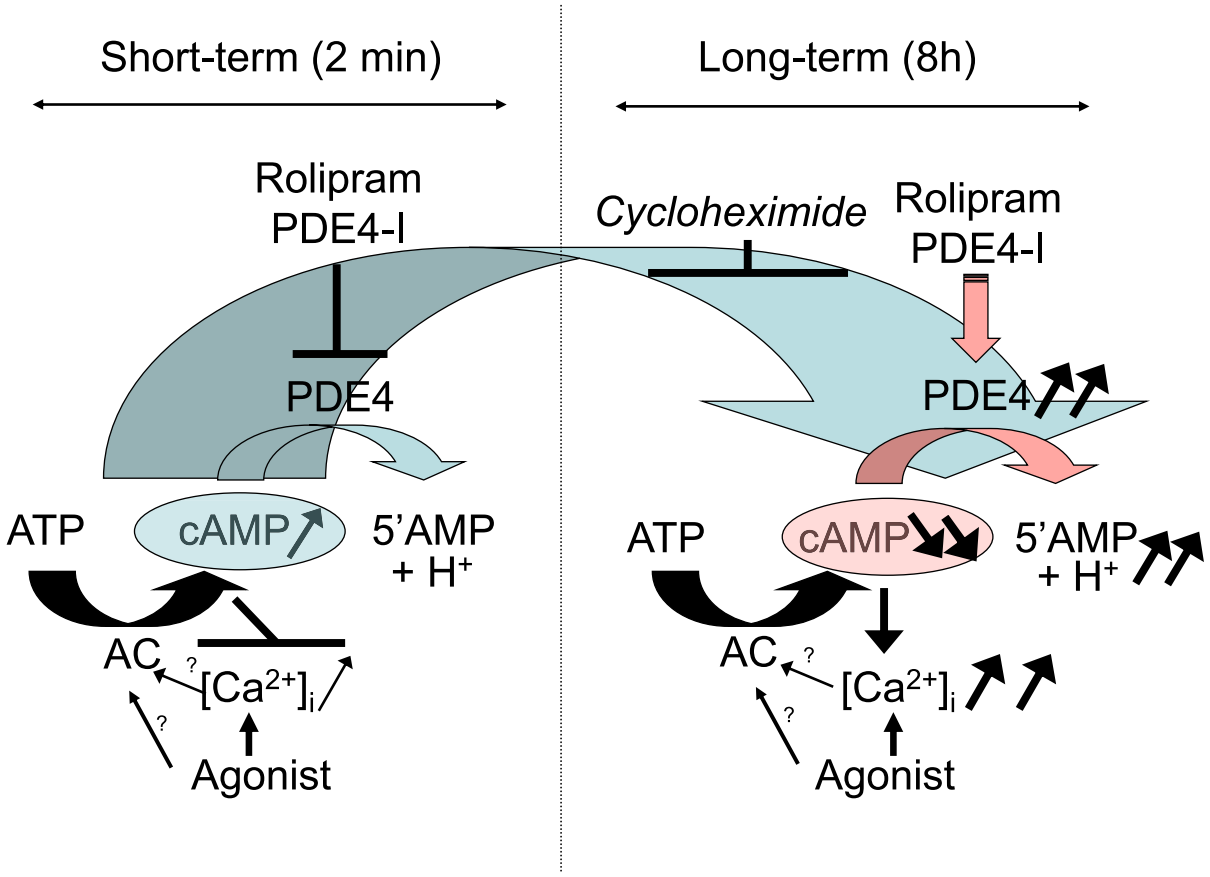

Fig. 3. Short- and long-term effect of rolipram and PDE4-inhibitor pre-treatment in HUVEC. Agonist induces $[Ca^{2+}]_I$ and may stimulate adenylyl cyclase (AC) activity. After 2 min pretreatment with cAMP elevating agents (rolipram, PDE4-I) enhances local increases of cAMP, reduces agonist stimulated $[Ca^{2+}]_I$, by inhibiting Ca^{2+}mobilization from internal stores. A sustained elevation of cAMP after 8 h pre-treatment with cAMP elevating agents, increases the expression and activity of PDE4 subtypes, which would speed up cAMP degradation.

include the activation of AT_1R, inducing a pulmonary "inflammatory storm" that can be counteracted by specific AT_1R antagonists which can be considered for treating COVID-19. However, this therapeutic approach might not be so effective in SARS-CoV-2 infection for CS.

Therefore, it would be better targeting selectively and potently PDE4 beyond AT_1R, as much as it is well established that PDE4 is up-regulated during inflammatory storm and that new potent and specific PDE4 inhibitors have been developed and marketed by pharmaceutical industries [9,10]. Importantly, losartan was previously shown to inhibit PDE4 activity, strengthening the possible PDE4 implication in COVID-19 [51]. Moreover, in that way, we showed that Ang-II up-regulates PDE4 activity in conjunction with PDE4A expression, supporting a new therapeutic approach [53]. Therefore, the use of specific PDE4 inhibitor should be adapted to selectively cure the pulmonary inflammation storm. In that way, studies performed on human pulmonary tissues [37,38] and PBMCs [49], clearly showed that inflammation up-regulates PDE4B, PDE4C, and subsequently increases TNF- α, IL-1 β, IL-6, IL-8, NFκB, p-p38-MAPK, mediators of inflammation. Interestingly, theses inflammatory processes have been overcome by a specific PDE4 inhibitor (Fig. 2).

Furthermore, knowing that PDE4 is up-regulated in CS [59], PDE4 inhibitor represents a promising treatment for COVID-19 CS, as much as a PDE4 inhibitor might be also active on virus replication and infection (see section **5**: PDE4 and viral infection). In accordance with our present suggestion, a treatment for COVID-19 was proposed by combining antiviral and anti-inflammatory treatments [68]. On our way, a rationale for evaluating PDE4 inhibition for mitigating against severe inflammation in COVID-19 pneumonia was lastly reported [69], as well as suggested in a commentary as a potential adjunct treatment targeting the cytokine storm in COVID-19 [70]. It could not be neglected that previously a company was developing a novel drug candidate against HIV-1 which presents both anti-inflammatory and anti-viral properties, notably by generating miR-124 [71].

Therefore, this review illustrates a great link between PDE4, and RAS in patients with CS, highlighting the potential role of these intricate pathways in the initiation and the propagation of SARS-CoV-2 and the subsequent CS-potentiation of acute lung injury. Besides, the modulation of PDE4 or RAS and CS in COVID-19, PDE4 inhibitors might be a promising way in the attenuation of COVID-19 induced-CS-ARDS.

7. Future directions

Altogether, beneath AT_1R stimulation, up-regulation of PDE4 is associated with H^+ production, inducing ALI, likely potentiated by CS. It should be pointed out that Raoult and colleagues have prone against SARS-CoV-2 the use chloroquine, which mediates anti-inflammatory response, interfere with the pH-dependent endosome-mediated viral entry of enveloped virus which is activated at acidic pH [72]. Interestingly, the use of PDE4 inhibitor might overcome the decrease in pH (see Fig. 2) and consequently might inhibit the release of the viral genome into the cytosol [72]. Interestingly, Kono et al. reported that chloroquine inhibits also the activation of active protein kinase p38-MAPK, which is involved in the multiplication of HCoV-229E [73], similarly to PDE4 inhibitors [46,49].

Therefore, other extensive studies, such as PDE4 implication, are recommended to explore the plausible scenario of CS and risk of COVID-19, as well as new opportunities for its treatment. In that way, it was reported on the 16th of June 2020, by Martin Landray, Professor of

Medicine and Epidemiology at the University of Oxford, that the low-cost dexamethasone reduces death by up to one third in hospitalized patients with severe respiratory complications of COVID-19 [74]. Thus, it must be pointed out that it was previously reported that PDE4 inhibition has an additive anti-inflammatory effect toward dexamethesone, related to its specific increased phospho-CREB nuclear translocation effect [75]. In the same field, it was hypothesized that pentoxifylline, might be used to treat COVID-19 as an immunomodulator and regulator of the renin-angiotensin system [76], it must be indicated that pentoxifylline inhibits PDEs [77] see Table 1. Lastly, the rational for PDE4 inhibitor uses in COVID-19 patients was pointed out in a general review on COVID-19 treatments [78].

In that context, it must be pointed out that not only PDE4 play a major role in lung, PDE4 also directly contributes in the regulation of endothelial cells [40,79], smooth muscle cells [7,37–39], and inflammatory cells such as PBMCs [49,60], to govern cell proliferation and inflammation, therefore contributing also by these ways in ARDS and ALI.

In endothelial cells, PDE4 is up-regulated in VEGF-induced angiogenesis [40,41], whereas PDE4 inhibitor, associated to PDE2 inhibitor, overcomes angiogenesis [41,80]. Thus, it was recently suggested to investigate whether anti-angiogenesis drugs in combination with other modalities could be considered in the treatment of COVID-19 [81]. Interestingly, PDE4 inhibitors were pointed out as a therapeutic approach to treat capillary leakage in systemic inflammation [82], also as therapy against eosinophil induced lung injury [83], attesting another way for PDE4 inhibitors to cure inflammation and intervene in COVID-19 treatment. Furthermore, PDE4 is also implicated in vascular smooth muscle cell proliferation, since in human pulmonary smooth muscle cells PDE4 inhibitors increase cAMP level and induce anti-proliferative effects [84]. However, it must be pointed out that long-term effect (>8h) of PDE4 inhibitors in rat smooth muscle cells [85], as well as in human endothelial cells [86], increases cAMP hydrolysis by PDE4, being in endothelial cells related to PDE4 up-regulated expressions. Therefore, it should be hypothesized that rapid treatment with PDE4 inhibitor might overcome COVID-19 aggression, whether late treatment by PDE4 inhibitor induces up-regulated PDE4 expression, increasing its activity, as does CS (Fig. 3). Hopefully, this increase in PDE4 activity might be overcome in the presence of its specific PDE4 inhibitor opening a new way for treating COVID-19 and CS.

CRediT authorship contribution statement

Claire Lugnier: Conceptualization, Writing - review & editing. **Hayder M. Al-Kuraishi:** Conceptualization, Writing - original draft. **Eric Rousseau:** Writing - review & editing.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgments

The authors would like to thank everyone working on the front lines of the COVID-19 pandemic, and make this work in memory to all people that suffers for this pandemic, losing their family members.

References

[1] J. Dyall, R. Gross, J. Kindrachuk, R.F. Johnson, G.G. Olinger, L.E. Hensley, et al., Middle East respiratory syndrome and severe acute respiratory syndrome: current therapeutic options and potential targets for novel therapies, Drugs 77 (18) (2017) 1935–1966.

[2] Q. Li, X. Guan, P. Wu, X. Wang, L. Zhou, Y. Tong, et al., Early transmission dynamics in Wuhan, China, of novel coronavirus–infected pneumonia, N. Engl. J. Med. 382 (13) (2020) 1199–1207.

[3] F. Zhou, T. Yu, R. Du, G. Fan, Y. Liu, Z. Liu, et al., Clinical course and risk factors for mortality of adult in patients with COVID-19 in Wuhan, China: a retrospective cohort study. Lancet 2020;395(10229):1054-1062, 2020.

[4] M. Shurin, A. Morris, A. Wells, S.E. Wheeler, Assessing immune response to SARS-CoV-2 infection, Immunotargets Ther. 9 (2020) 111–114.

[5] J. Yang, Y. Zheng, X. Gou, K. Pu, Z. Chen, Q. Guo, et al., Prevalence of comorbidities in the novel Wuhan coronavirus (COVID-19) infection: a systematic review and meta-analysis, Int. J. Infect. Dis. 94 (2020) 91–95.

[6] X. Yue, T.M. Basting, T.W. Flanagan, J. Xu, T.D. Lobell, N.W. Gilpin, et al., Nicotine downregulates the compensatory angiotensin-converting enzyme 2/angiotensin type 2 receptor of the renin–angiotensin system, Ann. Am. Thorac. Soc. 15 (2018) S126–S127.

[7] T. Keravis, C. Lugnier, Cyclic nucleotide phosphodiesterase (PDE) isozymes as targets of the intracellular signalling network: benefits of PDE Inhibitors in various diseases and perspectives for future therapeutic developments, Br. J. Pharmacol. 165 (5) (2012) 1288–1305.

[8] F. Ahmad, T. Murata, K. Shimizu, E. Degerman, D. Maurice, V. Manganiello, Cyclic nucleotide phosphodiesterases: important signaling modulators and therapeutic targets, Oral Dis. 21 (1) (2015) e25–e50.

[9] G.S. Baillie, G.S. Tejeda, M.S. Kelly, Therapeutic targeting of 3′,5′-cyclic inhibition and beyond, Nat. Rev. Drug Discov. 18 (10) (2019) 770–796.

[10] C. Lugnier, A. Meyer, S. Talha, B. Geny, Cyclic nucleotide phosphodiesterases: new targets in the metabolic syndrome? Pharmacol. Ther. 208 (2020), 107475.

[11] J.M. Oakes, R.M. Fuchs, J.D. Gardner, E. Lazartigues, X. Yue, Nicotine and the renin-angiotensin system, Am. J. Physiol. Regul. Integr. Comp. Physiol. 315 (5) (2018) R895–R906.

[12] K.K. Ng, J.R. Vane, Fate of angiotensin I in the circulation, Nature 218 (5137) (1968) 144–150.

[13] M.A. Parikh, C.P. Aaron, E.A. Hoffman, J.E. Schwartz, J. Madrigano, J.H.M. Austin, et al., Angiotensin-converting inhibitors and angiotensin II receptor blockers and longitudinal change in percent emphysema on computed tomography. The multi-ethnic study of atherosclerosis lung study, Ann. Am. Thorac. Soc. 14 (5) (2017) 649–658.

[14] H. Jia, Pulmonary angiotensin-converting enzyme 2 (ACE2) and inflammatory lung disease, Shock 46 (3) (2016) 239–248.

[15] C. Glynos, S.I. Bibli, P. Katsaounou, A. Pavlidou, C. Magkou, V. Karavana, S. Topouzis, I. Kalomenidis, Comparison of the effects of e-cigarette vapor with cigarette smoke on lung function and inflammation in mice, Am. J. Physiol. Lung Cell. Mol. Physiol. 315 (5) (2018) L662–L672, https://doi.org/10.1152/ajplung.00389.2017.

[16] Q. Wang, I.K. Sundar, D. Li, J.H. Lucas, T. Muthumalage, S.R. McDonough, I. Rahman, E-cigarette-induced pulmonary inflammation and dysregulated repair are mediated by nAChR α7 receptor: role of nAChR α7 in SARS-CoV-2 Covid-19 ACE2 receptor regulation, Respir. Res. 21 (1) (2020) 154, https://doi.org/10.1186/s12931-020-01396-y.

[17] J.P. Changeux, Z. Amoura, F.A. Rey, M. Miyara, A nicotinic hypothesis for Covid-19 with preventive and therapeutic implications, C R Biol. 343 (1) (2020) 33–39, https://doi.org/10.5802/crbiol.8.

[18] H.A. Tindle, P.A. Newhouse, M.S. Freiberg, Beyond smoking cessation: investigating medicinal nicotine to prevent and treat COVID-19, Nicotine Tob. Res. 22 (9) (2020) 1669–1670, https://doi.org/10.1093/ntr/ntaa077.

[19] M. Conti, W. Richter, C. Mehats, G. Livera, J.Y. Park, C. Jin, Cyclic AMP-specific PDE4 phosphodiesterases as critical components of cyclic AMP signaling, J. Biol. Chem. 278 (8) (2003) 5493–5496.

[20] Q. Lu, E. Gottlieb, S. Rounds, Effects of cigarette smoke on pulmonary endothelial cells, Am. J. Physiol. Lung Cell. Mol. Physiol. 314 (5) (2018) L743–L756.

[21] Y. Gu, X.H. Yang, M.N. Dookhun, J.S. Zhou, S.L. Xia, H. Zhang, et al., Correlation between plasma-soluble angiotensin-converting enzyme 2, anti-angiotensin-converting enzyme 2, and angiotensin-(1–7) in patients with chronic heart failure, Cardiol. Plus 4 (1) (2019) 22.

[22] M. Podowski, C. Calvi, S. Metzger, K. Misono, H. Poonyagariyagorn, A. Lopez-Mercado, et al., Angiotensin receptor blockade attenuates cigarette smoke-induced lung injury and rescues lung architecture in mice, J. Clin. Invest. 122 (1) (2012) 229–240.

[23] I. Berlin, D. Thomas, A.L. Le Faou, J. Cornuz, COVID-19 and smoking [published online ahead of print, 2020 Apr 3]. Nicotine Tob Res. ntaa059. doi: 10.1093/ntr/ntaa059.

[24] W.J. Guan, Z.Y. Ni, Y. Hu, W.H. Liang, C.Q. Ou, J.X. He, et al., Clinical characteristics of coronavirus disease 2019 in China, N. Engl. J. Med. 382 (18) (2020) 1708–1720.

[25] Y. Bai, L. Yao, T. Wei, F. Tian, D.Y. Jin, L. Chen, et al., Presumed asymptomatic carrier transmission of COVID-19, JAMA 323 (14) (2020) 1406–1407.

[26] J.L. Ods, N. Kabbani, Is nicotine exposure linked to cardiopulmonary vulnerability to COVID-19 in the general population? FEBS J. (2020) https://doi.org/10.1111/febs.15303. 10.1111/febs.15303.

[27] X. Pang, Y. Cui, Y. Zhu, Recombinant human ACE2: potential therapeutics of SARS-CoV-2 infection and its complication, Acta Pharmacol. Sin. 41 (9) (2020) 1255–1257.

[28] Pena Silva RA. Cardiovascular oxidative stress: recent findings on ACE2 and MAO. 2012. doi: 10.17077/etd.109quuf9.

[29] Y.H. Hung, W.Y. Hsieh, J.S. Hsieh, F.C. Liu, L.C. Tsai, L.C. Lu, et al., Alternative roles of STAT3 and MAPK signaling pathways in the MMPs activation and

progression of lung injury induced by cigarette smoke exposure in ACE2 knockout mice, Int. J. Biol. Sci. 12 (2016) 454–465.
[30] P. Russo, S. Bonassi, R. Giacconi, M. Malavolta, C. Tomino, F. Maggi, COVID-19 and smoking: is nicotine the hidden link? Eur. Respir. J. 55 (6) (2020) 2001116.
[31] C.I. Vardavas, K. Nikitara, COVID-19 and smoking: a systematic review of the evidence, Tob. Induc. Dis. 18 (2020) 20.
[32] Q. Cui, C. Huang, X. Ji, W. Zhang, F. Zhang, L. Wang, Possible inhibitors of ACE2, the receptor of 2019-nCoV. 2020 Preprints 2020020047 (doi: 10.20944/preprints202002.0047.v1).
[33] J.S. Alqahtani, T. Oyelade, A.M. Aldhahir, S.M. Alghamdi, M. Almehmadi, A. S. Alqahtani, et al., Prevalence, severity and mortality associated with COPD and smoking in patients with COVID-19: a rapid systematic review and meta-analysis, PLoS ONE 15 (5) (2020), e0233147.
[34] H.M. Salah, Y. Sharma, J. Mehta, Smoking doubles the mortality risk in COVID-19: a meta-analysis of recent reports and potential mechanisms, Cureus 12 (10) (2020), e10837, https://doi.org/10.7759/cureus.10837.
[35] D. Simons, L. Shahab, J. Brown, O. Perski, The association of smoking status with SARS-CoV-2 infection, hospitalization and mortality from COVID-19: a living rapid evidence review with Bayesian meta-analyses (version 7). Addiction. 2020 Oct 2: 10.1111/add.15276. doi: 10.1111/add.15276.
[36] C. Lugnier, P. Schoeffter, A. Le Bec, E. Strouthou, J.C. Stoclet, Selective inhibition of cyclic nucleotide phosphodiesterases of human, bovine and rat aorta, Biochem. Pharmacol. 35 (10) (1986) 1743–1751.
[37] E. Rousseau, J. Gagnon, C. Lugnier, Biochemical and pharmacological characterization of cyclic nucleotide phosphodiesterase in airway epithelium, Mol. Cell. Biochem. 140 (1994) 171–175.
[38] E. Rousseau, J. Dostie, M. Taoudi-Benchekroun, A. Cadieux, C. Beaudry, C. Lugnier, Specific cyclic nucleotide phosphodiesterase inhibitors differently modulate contractile kinetics in airway smooth muscle, Can. J. Physiol. Pharmacol. 73 (12) (1995) 1784–1794.
[39] O. Pauvert, D. Salvail, E. Rousseau, C. Lugnier, R. Marthan, J.P. Savineau, Characterisation of cyclic nucleotide phosphodiesterase isoforms in the media layer of the main pulmonary artery, Biochem. Pharmacol. 63 (9) (2002) 1763–1772.
[40] L. Favot, T. Keravis, C. Lugnier, Modulation of VEGF-induced endothelial cell cycle protein expression through cyclic AMP hydrolysis by PDE2 and PDE4, Thromb. Haemost. 92 (2004) 634–645.
[41] T. Keravis, P.A. Silva, L. Favot, C. Lugnier, Role of PDEs in vascular health and disease : Endothelial PDEs and angiogenesis. In: JA Beavo, SH Francis, & MD Houslay (Eds.). Cyclic Nucleotide Phosphodiesterases in Health and Disease. (pp.419-439). Boca Raton: CRC Press.
[42] H. Li, J. Zuo, W. Tang, Phosphodiesterase-4 inhibitors for the treatment of inflammatory diseases, Front. Pharmacol. 9 (2018) 1048.
[43] J.J. Bourguignon, L. Désaubry, P. Raboisson, C.G. Wermuth, C. Lugnier, 9-Benzyladenines: potent and selective cAMP phosphodiesterase inhibitors, J. Med. Chem. 40 (1997) 1768–1770.
[44] E. Boichot, J.L. Wallace, N. Germain, M. Corbel, C. Lugnier, V. Lagente, J. J. Bourguignon, Anti-inflammatory activities of a new series of selective phosphodiesterase 4 inhibitors derived from 9-benzyladenine, J. Pharmacol. Exp. Ther. 292 (2000) 647–653.
[45] T. Keravis, F. Monneaux, I. Yougbaré, L. Gazi, J.J. Bourguignon, S. Muller, et al., Disease progression in MRL/lpr lupus-prone mice is reduced by NCS 613, a specific cyclic nucleotide phosphodiesterase type 4 (PDE4) inhibitor, PLoS ONE 7 (1) (2012), e28899.
[46] I. Yougbare, C. Morin, F.Y. Senouvo, C. Sirois, R. Albadine, C. Lugnier, et al., NCS 613, a potent and specific PDE4 inhibitor, displays anti-inflammatory effects on human lung tissues, Am. J. Physiol. Lung Cell. Mol. Physiol. 301 (4) (2011) L441–L450.
[47] I. Yougbare, L. Belemnaba, C. Morin, A. Abusnina, Y.F. Senouvo, T. Keravis, et al., NCS 613, a potent PDE4 inhibitor, displays anti-inflammatory and anti-proliferative properties on A549 lung epithelial cells and human lung adenocarcinoma explants, Front. Pharmacol. 11 (2020) 1266, https://doi.org/10.3389/fphar.2020.01266. eCollection 2020.
[48] I. Yougbare, T. Keravis, C. Lugnier, NCS 613, a PDE4 inhibitor, by increasing cAMP level suppresses systemic inflammation and immune complexes deposition in kidney of MRL/lpr lupus- prone mice. Biochim Biophys Acta Mol Basis Dis. 2020; 166019. doi: 10.1016/j.bbadis.2020.166019. Online ahead of print. PMID: 33227426.
[49] I. Yougbaré, G. Boire, M. Roy, C. Lugnier, E. Rousseau, NCS 613 exhibits anti-inflammatory effects on PBMCs from lupus patients by inhibiting p38 MAPK and NF-κB signalling pathways while reducing proinflammatory cytokine production, Can. J. Physiol. Pharmacol. 91 (5) (2013) 353–361.
[50] G. Chen, D. Wu, W. Guo, Y. Cao, D. Huang, H. Wang, et al., Clinical and immunological features of severe and moderate coronavirus disease 2019, J. Clin. Invest. 130 (5) (2020) 2620–2629.
[51] V. Segarra, M.I. Crespo, F. Pujol, J. Belata, T. Doménech, M. Miralpeix, et al., Phosphodiesterase inhibitory properties of losartan. Design and synthesis of new lead compounds, Bioorg. Med. Chem. Lett. 8 (5) (1998) 505–510.
[52] A.G. Teschemacher, E.P. Seward, Bidirectional modulation of exocytosis by angiotensin II involves multiple G-protein-regulated transduction pathways in chromaffin cells, J. Biol. Chem. 278 (8) (2003) 5493–5496.
[53] W. Mokni, T. Keravis, N. Etienne-Selloum, A. Walter, M.O. Kane, V.B. Schini-Kerth, et al., Concerted regulation of cGMP and cAMP phosphodiesterases in early cardiac hypertrophy induced by angiotensin II, PLoS ONE 5 (12) (2010), e14227.
[54] Y. Liang, J. Xie, D. Che, C. Zhang, Y. Lin, L. Feng, et al., MiR-124-3p helps to protect against acute respiratory distress syndrome by targeting p65 Biosci Rep. 2020;40(5):BSR20192132.
[55] W. Gao, H. Yang, MicroRNA-124-3p attenuates severe community–acquired pneumonia progression in macrophages by targeting tumor necrosis factor receptor–associated factor 6, Int. J. Mol. Med. 43 (2) (2019) 1003–1010, https://doi.org/10.3892/ijmm.2018.4011.
[56] S. Huang, X. Ge, J. Yu, Z. Han, Z. Yin, Y. Li, et al., Increased miR-124-3p in microglial exosomes following traumatic brain injury inhibits neuronal inflammation and contributes to neurite outgrowth via their transfer into neurons, FASEB J. 32 (2018) 512–528.
[57] W. Gao, H. Yang, MicroRNA-124-3p attenuates severe community-acquired pneumonia progression in macrophages by targeting tumor necrosis factor receptor-associated factor 6, Int. J. Mol. Med. 43 (2019) 1003–1010.
[58] S.J. Brake, K. Barnsley, W. Lu, K.D. McAlinden, M.S. Eapen, S.S. Sohal, Smoking upregulates angiotensin-converting enzyme-2 receptor: a potential adhesion site for novel coronavirus SARS-CoV-2 (Covid-19), J. Clin. Med. 9 (2020) 841.
[59] R. Barber, G.S. Baillie, R. Bergmann, M.C. Shepherd, Differential expression of PDE4 cAMP phosphodiesterase isoforms in inflammatory cells of smokers with COPD, smokers without COPD, and nonsmokers, Am. J. Physiol. Lung Cell. Mol. Physiol. 287 (2) (2004) L332–L343.
[60] H. Zuo, B. Han, W.J. Poppinga, L. Ringnalda, L.E.M. Kistemaker, A.J. Halayko, et al., Cigarette smoke up-regulates PDE3 and PDE4 to decrease cAMP in airway cells, Br. J. Pharmacol. 175 (14) (2018) 2988–3006, https://doi.org/10.1111/bph.14347. Epub 2018 Jun 3.
[61] J.B. Angel, B.M. Saget, S.P. Walsh, T.F. Greten, C.A. Dinarello, P.R. Skolnik, et al., Rolipram, a specific type IV phosphodiesterase inhibitor, is a potent inhibitor of HIV-1 replication, AIDS 9 (10) (1995) 1137–1144.
[62] J. Navarro, C. Punzon, J.L. Jimenez, E. Fernandez-Cruz, A. Pizarro, M. Fresno, et al., Inhibition of phosphodiesterase type IV suppresses human immunodeficiency virus type 1 replication and cytokine production in primary T cells: involvement of NF-κB and NFAT, J. Virol. 72 (1998) 4712–4720.
[63] P. Secchiero, D. Zella, S. Curreli, P. Mirandola, S. Capitani, R.C. Gallo, et al., Pivotal role of cyclic nucleoside phosphodiesterase 4 in Tat-mediated $CD4^+$ T cell hyperactivation and HIV type 1 replication, PNAS 97 (26) (2000) 14620–14625.
[64] Y. Sun, L. Li, F. Lau, J.A. Beavo, E.A. Clark, Infection of CD4+ memory T cells by HIV-1 requires expression of phosphodiesterase 4, J. Immunol. 165 (4) (2000) 1755–1761.
[65] G. Li, J.I. Nunoya, L. Cheng, N. Reszka-Blanco, L.C. Tsao, J. Jeffrey, et al., Regulatory T Cells Contribute to HIV-1 Reservoir persistence in CD4+ T cells through cyclic adenosine monophosphate–dependent mechanisms in humanized mice in vivo, J Inf Dis. 216 (12) (2017) 1579–1591.
[66] R. Chemboli, R. Kapavarapu, K. Deepti, K.R.S. Prasad, A.G. Reddy, A.V.D. N. Kumar, et al., Pyrrolo[2,3-*b*]quinoxalines in attenuating cytokine storm in COVID-19: their sonochemical synthesis and *in silico / in vitro* assessment, J. Mol. Struct. 1230 (2021), 129868, https://doi.org/10.1016/j.molstruc.2020.129868.
[67] K. Kuba, Y. Imai, S. Rao, H. Gao, F. Guo, B. Guan, et al., A crucial role of angiotensin converting enzyme 2 (ACE2) in SARS Coronavirus-Induced lung injury, Nat. Med. 11 (8) (2005) 875–879.
[68] J. Stebbing, A. Phelan, I. Griffin, C. Tucker, O. Oechsle, D. Smith, et al., COVID-19: combining antiviral and anti-inflammatory treatments, Lancet Infect. Dis. 20 (2020) 400–402.
[69] C. Bridgewood, G. Damiani, K. Sharif, A. Watad, N.L. Bragazzi, L. Quartuccio, et al., Rationale for evaluating PDE4 inhibition for mitigating against severe inflammation in COVID-19 pneumonia and beyond, Isr. Med. Assoc. J. 22 (6) (2020) 335–339.
[70] M. Dalamaga, I. Karampela, C.S. Mantzoros, Commentary: Phosphodiesterase 4 inhibitors as potential adjunct treatment targeting the cytokine storm in COVID-19, Metab. Clin. Exper. 109 (2020), 154282.
[71] A. Vautrin, L. Manchon, A. Garcel, N. Campos, L. Lapasset, A.M. Laaref, et al., Both anti-inflammatory and antiviral properties of novel drug candidate ABX464 are mediated by modulation of RNA splicing, Sci. Rep. 9 (1) (2019) 792.
[72] C.A. Devaux, J.M. Rolain, P. Colson, D. Raoult, New insights on the antiviral effects of chloroquine against coronavirus: what to expect for COVID-19? Int. J. Antimicrob. Agents 55 (5) (2020), 105938 https://doi.org/10.1016/j.ijantimicag.2020.105938.
[73] M. Kono, K. Tatsumi, A.M. Imai, K. Saito, T. Kuriyama, H. Shirasawa, Inhibition of human coronavirus 229E infection in human epithelial lung cells (L132) by human coronavirus 229E infection in human epithelial lung cells (L132) by chloroquine: involvement of p38 MAPK and ERK. Antivir. Res. 20008; 77. doi: 10.1016/j.antiviral.2007.10.011.
[74] https://www.ox.ac.uk/news/2020-06-16-low-cost-dexamethasone-reduces-death-one-third-hospitalised-patients-severe.
[75] S. Grundy, J. Plumb, M. Kaur, D. Ray, D. Singh, Additive anti-inflammatory effects of corticosteroids and phosphodiesterase-4 inhibitors in COPD CD8 cells, Respir. Res. 17 (2016) 9.
[76] V. Maldonado, J.C. Alderete, Repositioning of pentoxifylline as an immunomodulator and regulator of the renin-angiotensin system in the treatment of COVID-19, Med. Hypotheses 9 (2020), 109988.
[77] L. Tetsi, A.L. Charles, S. Paradis, A. Lejay, S. Talha, B. Geny, C. Lugnier, Effects of cyclic nucleotide phosphodiesterases (PDEs) on mitochondrial skeletal muscle functions, Cell. Mol. Life Sci. 74 (10) (2017) 1883–1893, https://doi.org/10.1007/s00018-016-2446-0.
[78] M. Giorgi, S. Cardarelli, F. Ragusa, M. Saliola, S. Biagioni, G. Poiana, et al., Phosphodiesterase inhibitors: could they be beneficial for the treatment of COVID-19? Int. J. Mol. Sci. 21 (15) (2020) 5338, https://doi.org/10.3390/ijms21155338.

[79] C. Lugnier, V.B. Schini, Characterization of cyclic nucleotide phosphodiesterases from cultured bovine aortic endothelial cells, Biochem. Pharmacol. 39 (1) (1990) 75–84.
[80] L. Favot, T. Keravis, V. Holl, A. Le Bec, C. Lugnier, VEGF-induced HUVEC migration and proliferation are decreased by PDE2 and PDE4 inhibitors, Thromb. Haemost. 90 (2) (2003) 334–343.
[81] R. Amraei, N. Rahimi, COVID-19, renin-angiotensin system and endothelial dysfunction, Cells 9 (7) (2020) 1652, https://doi.org/10.3390/cells9071652.
[82] M.A. Schick, C. Wunder, J. Wollborn, N. Roewer, J. Waschke, C.T. Germer, et al., Phosphodiesterase-4 inhibition as a therapeutic approach to treat capillary leakage in systemic inflammation, J. Physiol. 590 (11) (2012) 2693–2708.
[83] T.J. Torphy, M.S. Barnette, D.W. Hay, D.C. Underwood, Phosphodiesterase IV inhibitors as therapy for eosinophil-induced lung injury in asthma, Environ. Health Perspect. 102 Suppl 10 (Suppl 10) (1994) 79–84.
[84] E.J. Growcott, K.G. Spink, X. Ren, S. Afzal, K.H. Banner, J. Wharton, Phosphodiesterase type 4 expression and anti-proliferative effects in human pulmonary artery smooth muscle cells, Respir. Res. 7 (1) (2006) 9.
[85] R.J. Rose, H. Liu, D. Palmer, D.H. Maurice, Cyclic AMP-mediated regulation of vascular smooth muscle cell cyclic AMP phosphodiesterase activity, Br. J. Pharmacol. 122 (2) (1997) 233–240.
[86] M. Campos-Toimil, T. Keravis, F. Orallo, K. Takeda, C. Lugnier, Short-term or long-term treatments with a phosphodiesterase-4 (PDE4) inhibitor result in opposing agonist-induced $Ca^{(2+)}$ responses in endothelial cells, Br. J. Pharmacol. 154 (1) (2008) 82–92.
[87] A. Hatzelmann, C. Schudt, Anti-inflammatory and immunomodulatory potential of the novel PDE4 inhibitor roflumilast in vitro, J. Pharmacol. Exp. Ther. 297 (2001) 267–279.