RELATIONSHIPS BETWEEN CHRONIC OBSTRUCTIVE PULMONARY DISEASE 
AND LUNG CANCER: BIOLOGICAL INSIGHTS

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ABSTRACT

Lung cancer (LC) has become one of the leading causes of preventable death in the last few decades. Cigarette smoking (CS) stays as the main etiologic factor of LC despite that many other causes such as occupational exposures, air pollution, asbestos, or radiation have also been implicated. Patients with chronic obstructive pulmonary disease (COPD), which also represents a major cause of morbidity and mortality in developed countries, exhibit a significantly greater risk of LC. The study of the underlying biological mechanisms that may predispose patients with chronic respiratory diseases to a higher incidence of LC has also gained much attention in the last few years. The present review has been divided into three major sections in which different aspects have been addressed: 1) relevant etiologic agents of LC, 2) studies confirming the hypothesis that COPD patients are exposed to a greater risk of developing LC, and 3) evidence on the most relevant underlying biological mechanisms that support the links between COPD and LC. Several carcinogenic agents have been described in the last decades but CS remains to be the leading etiologic agent in most geographical regions in which the incidence of LC is very high. Growing evidence has put the line forward the implications of COPD and especially of emphysema in LC development. Hence, COPD represents a major risk factor of LC in patients. Different avenues of research have demonstrated the presence of relevant biological mechanisms that may predispose COPD patients to develop LC. Importantly, the so far identified biological mechanisms offer targets for the design of specific therapeutic strategies that will further the current treatment options for patients with LC. Prospective screening studies, in which patients with COPD should be followed up for several years will help identify biomarkers that may predict the risk of LC among these patients. Word count: 300
INTRODUCTION

Lung cancer (LC) has become one of the leading causes of preventable death in the last few decades (1). Cigarette smoking (CS) stands as the main etiologic factor of LC despite that many other causes such as occupational exposures, air pollution, asbestos, or radiation have also been implicated (1-6). In the 1950s and 1960s, several epidemiologic studies were conducted, in which the links between CS and LC were clearly established (1;7-9). On the other hand, the combination of CS with several environmental or occupational agents may increase the risk of LC in exposed individuals (1). Presently research on the epidemiology of LC is still very active as primary prevention continues to be the most relevant target. Moreover, indoor and outdoor pollutants, which may vary with time, and the components of CS (proportions of tar and nicotine), are also a matter of research nowadays (1). Additionally, the changes in the histopathological characteristics of LC in many developed countries, with a significant rise in the frequency of adenocarcinoma, have also prompted research in this field.

Molecular epidemiology focuses on the elucidation of the biological mechanisms that favor malignancy in the lung parenchyma and airways of smokers and on the factors that enhance susceptibility to LC. In line with this, it has also been well established that patients with underlying respiratory conditions such as chronic obstructive pulmonary disease (COPD), which also represents a major cause of morbidity and mortality in developed countries, exhibit a significantly greater risk of LC (3;10-17). Importantly, in patients with moderate-to-severe COPD, the prevalence of LC can go up as high as five-fold that of smokers without the disease (18-24). Furthermore, the epidemiologic relevance of emphysema in the development of LC in patients with COPD has also been highlighted (2-5;16). The need for well-validated and practical LC screening tools to be implemented in clinical settings has also been underscored in several recent studies (2-5;17).
The study of the underlying biological mechanisms that may predispose patients with chronic respiratory diseases to a higher incidence of LC has also gained much attention in the last few years by several investigators including those listed in this review (25-31). For instance, oxidative and nitrosative stress as a result of reactive oxygen and nitrogen species (ROS and RNS, respectively) were shown to favor carcinogenesis through the activation of cellular processes that result in neoplastic transformation or the induction of DNA mutations (32). Other investigations have also demonstrated the contribution of oxidative damage, inflammatory events, and tumor microenvironment to lung carcinogenesis in patients and animal models (19;20;25-31;33-37).

The present review has been divided into three different sections in which the main topics introduced herein have been reviewed. First of all, the most relevant etiologic agents of LC are being briefly described. Secondly, the different studies that have confirmed the underlying hypothesis that COPD patients are exposed to a greater risk of developing LC are also reviewed. Finally, in the third place, evidence on the most relevant underlying biological mechanisms that support the links between COPD and LC, which account for the greater predisposition of these patients to lung tumorigenesis, is also described in the current review.

MAIN ETIOLOGIC AGENTS OF LC

As abovementioned, in this century, CS still continues to be the most important etiologic factor of LC. Nonetheless, several other agents such as indoor and outdoor pollutants, which may act synergistically with CS, also play a relevant role in the etiology of LC. A brief description of the most relevant etiologic factors follows below (Figure 1).

Carcinogenic effects of CS

We need to go back in history as far as the 1950s in order to understand the etiology of LC. Indeed, the first epidemiologic studies that demonstrated the links between the deleterious
effects of CS on the airways and lungs of smokers and LC development were published by British and American scientists in the 1950s and 1960s (1;7-9). Importantly, the duration of smoking and the number of cigarettes smoked were shown to increase the risk of LC among the smokers (38;39). Specifically, in other seminal studies, the duration of CS predicted a higher risk of LC than the amount of cigarettes smoked daily (38;39). Thus, these findings have important implications on the age at which individuals start smoking, which is presently more common in younger adolescents.

The composition of cigarettes has also changed considerably throughout time. Currently, the use of filtered cigarettes is predominant over the unfiltered cigarettes that had been used in the last century. The proportions of nicotine and tar, which includes several types of cancer promoter chemicals in the condensable residue of CS, have varied throughout time. Studies have demonstrated that in smokers consuming filtered cigarettes with lower tar proportions the risk of LC was reduced compared to those exposed to unfiltered cigarettes with higher tar yields (40-42). In another investigation (43), in smokers consuming high-yield tar cigarettes the risk of LC was significantly greater than in low- and medium-yield smokers, suggesting that tar yields are a risk factor of LC. The risks for current and past-smokers have also been reported. As such, compared to lifelong non-smokers, the risk of LC was estimated to be four-fold higher among former smokers, while it was 14-fold greater among current smokers (44). The composition of chemicals included in the cigarette may also greatly account for the pattern changes in histological types of LC that have been observed in the last few decades. In this regard, a shift towards a predominance of adenocarcinoma over squamous and small cell carcinomas has been reported since late 1970s in several investigations (45-47). Moreover, evidence has also shown that passive smokers who are exposed to second-hand smoke have a higher risk of LC, especially that of nonsmoking women with a smoker husband (48;49).
Diet and physical activity

Importantly, factors such as the diet and physical activity also seem to play a role in the risk of LC in smokers. For instance, fruits, vegetables, and antioxidant micronutrients may exert protective effects against LC (50). Results obtained from case-control and cohort studies have demonstrated that smokers with an elevated daily intake of vegetables and fruits, especially the latter, had a lower risk of LC than those subjects who did not follow this type of diet (51-53). Moreover, diets rich in other types of nutrients that are abundantly present in tomatoes and cruciferous vegetables (cross-like shape as a result of the four equal-sized petals in their flowers) such as vitamins A, carotenoids, C, and folic acid, and fiber induced protective effects against LC in smokers compared to those who did not include this sort of elements in their diet (52;54-56). Despite that the studies aimed to evaluate the potential beneficial effects of certain nutrients in the reduction of LC risk in smokers are hard to conduct and interpret (confounding lifestyle factors associated with smoking), it is currently possible to conclude that vegetable consumption protects against LC (1). Finally, high levels of physical activity have also been shown to correlate with a lower risk of LC among active smokers than those with a more sedentary lifestyle, even after adjusting for CS (1). In this regard, pulmonary rehabilitation and exercise training programs for several weeks also induced beneficial effects in patients with LC who underwent thoracic surgery for the treatment of their lung neoplasm (57).

Occupational exposure

The contribution of occupational exposure to LC has been estimated to range from 9 to 15%, which is relatively low in industrialized regions when compared to the implications of CS (1). LC has been associated especially with exposure to the following agents: tar, soot, arsenic, chromium, nickel, and silica dust (1). Nonetheless, exposure to these agents has been well controlled in developed countries in the last decades (1). Additionally, in epidemiologic
studies, exposure to diesel exhaust was also demonstrated to induce LC in truck drivers (58;59), railroad workers (60), and operators of heavy construction equipment (61). More recently, exposure to new technologies included in the environment and workplace such as sandblasting jean workers and hydraulic fracturing (fracking) have introduced new hazards that may lead to the development of occupational diseases including LC (62).

Asbestos, which consists of fibers of silicate minerals, may also cause LC in exposed individuals such as coal miners (63). First evidence comes from studies conducted in the United Kingdom and United States of America, where textile (63) and insulation (64;65) workers exhibited a significantly higher risk of LC than non-exposed individuals. Importantly, asbestos-induced LC depends on the duration of the exposure and is significantly increased by the influence of CS, which may favor the retention of asbestos fibers in the lungs (66;67).

Radiation

Exposure to ionizing radiation has shown a strong association with LC (68). Despite that exposure to radiation produced by x-rays, gamma rays, neutrons, and radon was shown to cause LC, the levels required to induce lung carcinogenesis were significantly greater than those usually experienced by the general population (1). As such, among atomic bomb non-smoker survivors, the radiation-related risks for LC were similar to those estimated for other solid neoplasms: 0.9 with a female: male sex ratio of 1.6 (69). Radon is a chemical inert gas that occurs naturally as a decay product of radium. The decay products of radon emit alpha particles of high energy and mass that may damage nuclear DNA of cells in the lungs and airways. In fact, a very high risk of LC (40%) was observed in underground miners of uranium who had been exposed to radon chronically (70). In the study, the rates of deaths from LC were 70% and 39% for never-smokers and current smokers, respectively (70). Furthermore, in buildings, radon is ubiquitously distributed as it enters directly from the soil, and its concentrations may vary from room to room, depending on the level of ventilation.
The effects of indoor exposure to radon are significantly lower than those seen as a result of occupational exposure as in uranium miners (1,70). Recently, it has been demonstrated that residential radon may increase LC risk up to 30% among never-smokers (71,72). However, in the studies, the potential influence of environmental CS on the risk of lung cancer could not be ruled out (1,71,72). In conclusion, despite that indoor exposure to radon has been suggested to cause LC, these assumptions still need to be definitely confirmed in future epidemiologic studies (1,71,72).

Air pollution

The line has been recently put forward that air pollutants also contain carcinogens, which may favor LC development (1,73). Potential carcinogens include polycyclic aromatic hydrocarbons, arsenic, nickel, and chromium, which are all produced by the combustion of fossil fuels (1). Descriptive studies have highlighted a potential role of air pollution in LC development, especially in urbanized areas (1). In line with this, the relationships between long-term exposure to particulate matter < 10 micrometer in diameter (PM$_{10}$), sulfur dioxide, nitrogen dioxide and mortality of LC have been analyzed in a cohort of Northern China for several years (1998-2009) (74). The results were not conclusive as age, the assignment method for air pollution exposure, and smoking history influenced the analyses of the study results (74). In another study conducted in Tianjin (Northern China), exposure to high concentrations of polycyclic aromatic hydrocarbons induced a greater risk of LC among the elderly in a similar fashion for both men and women (75). On the other hand, ambient fine particulate matter (PM$_{2.5}$) has recently been shown to account for 32% of total reported deaths in the 74 leading cities of China (76). Specifically, 20% of the reported deaths were attributed to cardiovascular, respiratory and lung cancer conditions (76). The investigators concluded that in certain regions of China, PM$_{2.5}$ imposes significant health risks that are even greater.
than those so far attributed to CS, thus action plans for Air Pollution Prevention and Control should be enforced, at least in specific geographical areas (76).

Recently, strong associations between small cell LC and adenocarcinoma (hazard ratios: 1.53 and 1.44, respectively) and exposure to high levels of PM$_{2.5}$ have also been reported in a cohort of women of the Canadian Cancer Registry (77). In other studies, however, the effects of air pollution as a risk factor of LC were significantly attenuated after adjusting for several factors such as CS and occupational exposure, even if the influence of urbanization persisted (1;78). Other approaches have included the analyses of the effects of factories and smelters in populations residing nearby. However, the results were not entirely conclusive (1). As a summary from different reports (1), it is possible to conclude that 1-2% of LC can be attributed to air pollution, although these proportions may vary widely across geographical regions.

**SUSCEPTIBILITY TO LC: INFLUENCE OF COPD AND OTHER CHRONIC RESPIRATORY CONDITIONS**

**Evidence and epidemics**

Underlying chronic respiratory conditions such as COPD and lung fibrosis, especially pneumoconiosis (1), increase the susceptibility of the patients to LC. Importantly, COPD and chronic airway obstruction have long been associated with LC development (2-5;10-13;16;79). Interestingly, the prevalence of LC e has been consistently greater in men than in females. However, in the last decades, evidence has shown a rising incidence of LC in women probably as a result of the increased numbers of female smokers, who were diagnosed with LC (1;80). Furthermore, among never-smokers the prevalence of LC in females was also significantly higher than in men (81). Most of women were diagnosed at advanced stages of
tumor progression, even among never-smokers, who exhibited similar survival rates to those reported in smokers with LC (81).

Therefore, the deployment of screening programs in patients with underlying respiratory diseases has recently emerged as an actual medical need in Western and Eastern societies (2;3;82-84). Progress has been made in the identification of the best screening programs for the early diagnosis of LC. In this regard, low-dose computerized tomography (LDCT) has been proposed as a useful screening tool for LC (85). In keeping with, several authors (3;4;86;87) have demonstrated the benefits of LDCT in the early diagnosis of LC among smokers. Nonetheless, other authors have claimed that in LC screening programs, LDCT should not be still widely applied in the general population up until more convincing results are published (2).

Interestingly, in a LC screening program using LDCT that was conducted in a region of Spain, the presence of COPD and especially of emphysema were strong predictors of LC among the study patients (3). The authors concluded that the results obtained in the LC screening program were comparable to observations that had been previously reported in other European programs, and that LDCT was a valid and feasible diagnostic tool in this context (3). In another study, the same investigators (4) showed that LC screening programs that are exclusively based on the National Lung Screening Trial (NLST) criteria may fail to identify all the cases of LC. On this basis, the investigators suggested that the application of the NLST criteria in patients with emphysema enhanced the detection rates of LC, while lowered the number of missed cases (4). Interestingly, the use of another diagnostic/therapeutic tool such as the endobronchial insertion of one-way valves for the treatment of severe dyspnea in patients with emphysema allowed for the early diagnosis of LC, especially during follow-up (82).
Risk factors of LC

With the aim to identify potential risk factors that may help predict LC morbidity and mortality other approaches have also been used in clinical settings. For instance, pleural and vascular invasion were demonstrated to influence survival and increase the risk of death in patients with non-small cell lung cancer (NSCLC) of small sizes, thus they could be used in a predictive risk model (5;84). Other observations have underscored that in patients with early stages, pneumonectomy rather than lobectomy was associated with poorer survival (12;13), and that airway obstruction was an important predictive factor to define 30-day mortality after lung surgery even in LC patients with advanced age (12;88).

Whether long-lasting effects of CS may be found in patients already diagnosed with LC has also been the matter of recent research. In this regard, in long-term survivor patients, recurrence and the appearance of second tumors were observed in the lungs and other organs of patients with LC as early as three years after the diagnosis (12;89). As the new tumors were also related to CS, the authors concluded that the multiple carcinogenic effects of CS persist several years after the LC diagnosis in patients with long survival (12;89). Thus, novel diagnostic tools are required in order to identify patients who may be at a higher risk of LC, with a special focus on patients with a long smoking history and/or the presence of underlying respiratory conditions such as COPD (90-93).

Interestingly, the risk factors of LC hospitalization have also been recently analyzed from the National Hospital Discharge Database in Spain (79). The conclusions from the study were that age and sex influenced the incidence of hospitalization in patients with LC and that while it decreased in men, a rise was observed in women, which was partly related to the presence of comorbidities (79). In keeping with, recent results obtained from the Spanish National Statistics Institute have shown that the age-adjusted mortality rates increased among Spanish women, while they decreased in men (94). These findings were related to a rise in the
prevalence of CS reported among women in Spain (94). Similar findings are probably expected in other geographical regions and deserve special attention (95;96). As a matter of fact, the incidence of CS and exposure to other carcinogenic agents represent major targets for LC prevention campaigns in Western and Eastern societies.

POTENTIAL BIOLOGICAL MECHANISMS MEDIATING LC DEVELOPMENT IN PATIENTS WITH COPD

Chronic inflammation

Chronic inflammation through the induction of several interleukins (IL) and cyclooxygenase-2 activity may be an important player in the lung tumor formation among patients with COPD (Figure 2) (97-100). Free radicals and proteases released by activated leukocytes together with the formation of tertiary lymphoid aggregates may conform the first step in tumor development of patients bearing underlying lung inflammatory conditions such as COPD (100). Moreover, these inflammatory molecules may interfere with key regulatory mechanisms such as cell death (apoptosis), autophagy, cell repair, and angiogenesis, which contribute to the neoproliferative process (97;98). Recently, migration (a key process in tumor progression) of NSCLC cells A549 was significantly increased in a chemotactic gradient produced by the serum of COPD patients compared to that of the healthy controls (29). In the serum of the patients, the concentrations of CCL21 and CXCL12, but not those of CXCL5, were significantly greater than levels found in the control subjects (29). Interestingly, the blockade of CCL21 and CXCL12 activities showed that the greater migration of the A549 cells observed in COPD was mediated by the former cytokine (CCL21) (29). The investigators concluded that CCL21 may favor cancer cell migration in the lungs of patients with COPD (29). As these results may offer an interesting therapeutic strategy to combat LC,
future studies should be specifically designed in order to demonstrate this mechanism in actual tumors from COPD patients.

Other cytokines and growth factors such as tumor necrosis factor (TNF)-alpha, vascular endothelial growth factor (VEGF), and transforming growth factor (TGF)-beta have also been shown to participate in the development of LC in patients with underlying respiratory conditions (30;101). Identification of additional inflammatory molecules that may be involved in the development of LC among patients with COPD and/or tumor progression is of relevance as they offer potential for the design of novel therapeutic strategies in the treatment of LC (100;102).

Cytokines released by T cells may play different roles in tumorigenesis. In line with this, Th1 lymphocytes, which release TNF-alpha, IL-2, and interferon-gamma, have been shown to exert antitumor effects, while Th2 cells, which mainly produce IL-4, predominantly favor tumor growth by inhibiting the host immune system (103). Importantly, LC relapse may also rely on alterations in the balance between Th1 and Th2 cytokines in patients (104-106). For instance, a rise in the systemic levels of Th2 cytokines was observed in patients with LC, while those of Th1 cytokines were decreased (106). Interestingly, after surgical resection of the lung tumor, levels of Th1 and Th2 cytokines were modified in the same patients (106). Specifically, blood levels of both IL-10 and IL-4 were significantly reduced after tumor resection compared to baseline levels before the surgery, despite that they remained significantly greater than those detected in the healthy control group of subjects (only baseline measurements) (106). A relationship between accumulation of myeloid-derived suppressor cells in patients with COPD and LC development has also been recently suggested (107). Recently, associations have also been reported between the levels of certain immune regulators in the bronchoalveolar lavage and disease progression such as metastasis and body weight loss in patients (108).
In a recent investigation from our group (unpublished observations), levels of Th1 cytokines were significantly greater in the tumors of patients with LC and underlying COPD than in those without the chronic respiratory condition. We concluded from these findings that patients with COPD may be somehow protected against tumor development and progression by the release of Th1 cytokines. Studies underway will shed light into the potential mechanisms linking the release of cytokines, chronic inflammation and the lung tumorigenesis in patients with underlying chronic respiratory conditions.

Type 1 (M1) and type 2 (M2) polarized macrophage subtypes play a significant role in tumorigenesis through the regulation of several functions such as cell adhesion, apoptosis, and senescence (104;105;109). Furthermore, macrophages may exert proinflammatory or anti-inflammatory functions depending on the secreted cytokines. In tumors, macrophages are the predominant cells within the inflammatory infiltrates. Importantly, M1 macrophages favor inflammation, whereas M2 macrophages promote anti-inflammatory actions and tissue repair. While M1 cells fight against tumor development, M2 macrophages exert the opposite effects, by promoting cancer growth, survival, progression, and dissemination (110). Recently, the profile of macrophages has been analyzed in the airways of COPD (111). As such, patients with COPD and no LC, airway macrophages did not follow the classic M1/M2 pattern, as a skewed transcriptomic profile that favors M2 macrophages was actually found (111). The authors concluded that this profile might favor tumorigenesis in COPD.

Recent unpublished observations from our group have also shown that in patients with LC, the number of M1 macrophages was reduced while a rise in M2 macrophages was observed in the same specimens. Additionally, a significantly greater M1/M2 ratio was detected in the tumors of LC patients with underlying COPD than in those of patients without this disease. These findings suggest that the prognosis of LC patients with underlying COPD
may be better than in those with no COPD. Nonetheless, further research is needed in order to confirm this hypothesis.

The implications of tumor microenvironment are also relevant in the study of the underlying biology that accounts for the greater predisposition of COPD patients to develop lung tumors. In this regard, tumor microenvironment induces immune suppression, reduces the efficacy of chemotherapy, and favors epithelial-to-mesenchymal transition (EMT) in the airways (type-II EMT, obliteration of small airways), which has recently emerged as a novel target for LC treatment (112). In line with this, a recent investigation showed that treatment of lung epithelial cells with CS extracts induced alveolar EMT through a cascade of biological mechanisms characterized by a rise in TGF-beta and Rac1/Smad2 signaling pathway (113). Interestingly, the blockade of TGF-beta activity attenuated EMT expression markers (113). The authors also concluded that these results may open a new avenue for research in the treatment of patients with LC and underlying COPD (113) as type II-EMT and angiogenesis (type-III EMT) favor lung tumor development (31).

Redox balance

Oxidative stress, defined as the imbalance between oxidants and antioxidants in favor of the former, represents another relevant contributing factor to LC progression (18-22;25-28;33;36) (Figure 3). Oxidative and nitrosative stress were shown to favor carcinogenesis through the activation of cellular processes that result in neoplastic transformation, the induction of DNA mutations (32;114), or even through induction of macrophage dysfunction (alterations in phagocytosis) (27). A rise in oxidant production was observed in several tissues of LC patients and in smokers (18;21;22;33). Proteins and DNA are major cellular target components for the action of oxidants that escape the cellular antioxidant systems. In this regard, several plasma proteins were strongly nitrated and oxidized in LC patients (20). Proteins involved in glycolysis, oxidant scavenging, and cellular structure were more severely...
nitrated in the lung tumor tissue compared to the non-tumor parenchyma in LC patients in another study (19). More recently, patients with advanced LC exhibited increased systemic oxidative stress levels compared to healthy controls (18;33).

In a previous study from our group (25), protein carbonylation levels, as measured by malondialdehyde (MDA)-protein adducts, were also increased in the normal epithelium of patients with LC, especially in patients with underlying COPD, whose levels were significantly greater than in those without this disease. These findings were consistent with those reported in previous investigations, in which a rise in different redox markers was demonstrated in lung tissues or blood of patients with LC (19;20;25;37;101).

Several structural and functional proteins were significantly more oxidized in the lung tumors and non-tumor parenchyma in patients with LC (26). In fact, proteins such as cofilin (34), vimentin (115), and alpha-1-antitrypsin (116;117) were also shown to be more oxidized in the normal epithelium of the airways distant to the neoplasm in patients with LC (25), and their function was altered as a result of the oxidative posttranslational modifications, which may contribute to lung destruction and emphysema (116;117).

On the other hand, several proteins including vimentin, actin, and carbonic anhydrase-1 were also identified to be tyrosine nitrated in the lung tumors of patients (19). Whether these patients might also have had underlying COPD was not analyzed in that investigation (19). However, no conclusive results have been recently encountered in a study conducted in our group as protein tyrosine nitration levels did not significantly differ in the lung tumors of patients with and without COPD (19).

In a recent study (26), a significant rise in mitochondrial superoxide dismutase (SOD)2 protein content was observed in the tumors compared to non-tumor parenchyma in both groups of patients. Additionally, when patients with LC and COPD were analyzed separately on the basis of their smoking history, the heaviest smokers were those exhibiting the actual
increase in SOD2 levels in the lung tumors (26). These findings suggest that chronic CS may further enhance the rise in SOD2 content observed in the tumors of those patients. The conclusions from these findings were that SOD2 may be a key survival mechanism for the cancer cells to proliferate in the tumors. In fact, another investigation (34) demonstrated that inhibition of SOD activity reduced tumor burden in mice and promoted cell death in several NSCLC lines of cells. Furthermore, SOD2 was also shown to favor cell migration and invasiveness of tumors in other investigations (117;118). More recently, mRNA and protein levels of SOD2 were also significantly increased in lung tumors and other cancer types in patients (37). The authors concluded that SOD2 may even be considered as a biomarker for cancer progression, from tumor growth to metastasis. Hence, SOD2 overexpression seems to be involved in tumorigenesis in patients with LC, particularly in those with COPD who have a history of CS. Thus, drugs targeted to block SOD2 activity could be of interest in clinical settings.

Protein levels of SOD1 were in general much greater in the patients with underlying COPD in both tumor and non-tumor lung specimens than in LC patients with no COPD (26). Interestingly, CS did not influence SOD1 levels in any of the LC patients with COPD. The conclusions were that SOD1 seems to participate in antioxidant defense of the lungs in COPD patients regardless of the presence of LC rather than in carcinogenesis (26).

The antioxidant enzyme catalase catalyzes the decomposition of hydrogen peroxide to water and oxygen, thus protecting the cells from oxidative damage. Importantly (26), catalase protein levels were significantly reduced in the tumors compared to non-tumor parenchyma in LC patients with and without COPD. Furthermore, in LC patients with COPD, the heaviest smokers were those showing the decrease in catalase levels in the tumor lesions compared to the non-tumor lungs (26). These findings are consistent with other studies in which catalase deficiency was shown to contribute to mammary tumorigenesis in rodents (119) and cancer in
patients (37). Collectively, it would be possible to conclude that catalase depletion seems to be involved in cancer development, especially in LC patients with underlying COPD, especially in those who were heavy smokers.

Systemic levels of the oxygen radical superoxide anion and oxidative stress markers were significantly greater, while blood levels of the antioxidant glutathione were reduced in the LC patients with COPD compared to those without this disease (26). Importantly, no significant differences between moderate and heavy smokers were seen in any of the markers analyzed in the blood samples when LC patients with COPD were further subdivided according to their smoking history. In this investigation (26), it was suggested that underlying COPD itself rather than chronic CS may account for the differential pattern of redox balance expression observed in the lung tumors and especially in the blood compartment of LC patients with and without COPD (26).

Glutathione play a relevant role in the detoxification of carcinogens and polycyclic aromatic hydrocarbons (120). Several mutations or deletions of glutathione transferases have been shown to increase susceptibility to develop cancers in patients (121;122). A meta-analysis showed that a specific genotype of glutathione transferases increased the risk to develop LC in Asian populations (120). Levels of the antioxidant reduced glutathione (GSH) were significantly lower in the plasma of LC patients with COPD than in patients without this disease. Interestingly, smoking history did not influence the results encountered in LC patients with underlying COPD, since no differences were detected between moderate and heavy smokers (26). Collectively, all these findings suggest that oxidative damage and antioxidant depletion may contribute to a greater risk to lung carcinogenesis, especially in patients with underlying COPD. Systemic levels of superoxide anion, protein carbonyls, GSH, and nitrotyrosine above a specific threshold levels were predictive of the presence of underlying COPD among the study patients with LC (26). The conclusions were that underlying COPD
may predispose patients to a higher risk to develop LC through the induction of increased levels of oxidative stress (26).

Collectively, oxidative stress appears to be a potential therapeutic target for the treatment of LC as antioxidants have recently demonstrated to exert antitumor effects by downregulating proliferating signaling pathways in experimental models (123;124). Furthermore, in the THP-1 cell line (macrophages), the antioxidant thymoquinone also attenuated the phagocytic alterations induced by CS exposure and lipopolysaccharide (27). Nonetheless, in another experimental mouse model of LC, the antioxidant N-acetyl cysteine did not show any beneficial effects on tumor growth (36). Taken together, these results suggest that more research is needed before antioxidants can be included in LC guidelines as potential therapies in actual patients.

Conclusions

Several carcinogenic agents have been described in the last decades but CS remains to be the leading etiologic agent in most of the geographical regions in which the incidence of LC is very high. Growing evidence has put the line forward the implications of COPD and especially of emphysema in LC development. Hence, COPD represents a major risk factor of LC in patients. Different avenues of research have demonstrated the presence of relevant biological mechanisms that may predispose COPD patients to develop LC. Importantly, the so far identified biological mechanisms offer targets for the design of specific therapeutic strategies that will further the current treatment options for patients with LC. Prospective screening studies, in which patients with COPD should be followed up for several years will help identify biomarkers that may predict the risk of LC among these patients.
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All authors declare that the manuscript has not been submitted elsewhere, that they took a significant part in the work and have approved the final version, that they have complied with the ethical standards, and that they agree AME publishing company, to get a license to publish the accepted article when the manuscript is accepted.

AUTHORS’ CONTRIBUTIONS

EB, VB, VC, JG, JLC, XM: Conception and design

EB, JLC, XM: collection and assembly of data

EB: Manuscript writing and revised final version

EB, VB, VC, JG, JLC, XM: Intellectual input to written draft

EB, VB, VC, JG, JLC, XM: Final approval of manuscript

AUTHOR DISCLOSURE STATEMENT

The authors declare that they have no competing interests in relation to the contents of this review article.
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FIGURE LEGENDS

**Figure 1**: Schematic representation of the most relevant etiologic factors of lung cancer in patients. *Cigarette smoking, occupational exposure (including several procedures and materials), diet, physical activity, radiation, and air pollution are the most relevant etiologic agents of lung cancer.*

**Figure 2**: Schematic representation of the potential role of cytokines in tumor development in patients with underlying COPD. *Cigarette smoking induces chronic inflammatory events characterized by the induction of several interleukins (IL), cyclooxygenase-2 activity, and cytokines. These inflammatory molecules interfere with key regulatory mechanisms such as cell death (apoptosis), cell repair, and angiogenesis, which contribute to the neoproliferative including tumor growth and metastasis.*

**Figure 3**: Schematic representation whereby redox imbalance may induce damage in cells. *Reactive oxygen species (ROS) are formed by the addition of electrons to the oxygen molecule leading to the formation of different ROS. Oxidative stress takes place in cells and tissues as a result of an imbalance between oxidants and antioxidants in favor of the former. Oxidative damage in tissues is induced through several mechanisms such as peroxidation of membrane lipids, alterations in nuclear DNA, or oxidation of cell proteins.*
ETIOLOGIC FACTORS OF LUNG CANCER

- Cigarette smoke
- Occupational exposure:
  - Tar
  - Soot
  - Arsenic
  - Chromium
  - Nickel
  - Silica dust
  - Diesel exhaust
  - Sandblasting
  - Hydraulic fracturing (fracking)
- Diet
- Physical activity
- Radiation
- Air pollution

Barreiro et al. Figure 1
INFLAMMATION

Cigarette smoking

↑ Cytokines: IL-1beta, Th-1 cytokines

↑ COX-2 activity

↑ Cytokines: IL-6, IL-8, IL-10

↓ Apoptosis

↓ Cell repair

↑ Angiogenesis

↑ Tumor growth & Metastasis

Barreiro et al. Figure 2
REACTIVE OXYGEN SPECIES

Addition of a single electron to the oxygen molecule through a reduction process leads to the sequential production of a series of reactive molecules such as $\text{O}_2^\cdot$, $\text{H}_2\text{O}_2$, and $^\cdot\text{OH}$.

OXIDATIVE STRESS

Greater levels of ROS production than those normally neutralized by intracellular antioxidant defenses

Oxidative damage to other cellular components of the cell:
- peroxidation of membrane phospholipids
- modification of nuclear DNA
- alterations in proteins

Barreiro et al. Figure 3
LETTER TO THE EDITOR

Manuscript ID: JTD-16-1632

Title: RELATIONSHIPS BETWEEN CHRONIC OBSTRUCTIVE PULMONARY DISEASE AND LUNG CANCER

Dear Dr. Barreiro,

C1

Thank you very much for submitting the above article to Journal of Thoracic Disease. Your submission has been assessed by reviewers and discussed by the editorial team. We have found your manuscript potentially acceptable for publication. However, the manuscript will require major revision based on the reviewers’ comments, which is attached.

In order to expedite the processing of the revised manuscript, please be as specific as possible in your response to the reviewer(s). The revised manuscript (keep the track changes) and your reply to the comments will be forwarded to the referees for re-review. Please give your response to the comments point-by-point as shown in the following format.

Comment 1: **********
Reply 1: **********

Comment 2: **********
Reply 2: **********

R1

We thank the Editor for having given us the opportunity to revise our manuscript according to the reviewers’ comments. We have paid attention to all the reviewers’ concerns and have modified our manuscript accordingly. Moreover, responses to each of the reviewers’ comments have also been provided in a separate document.

Additionally, we would like to underscore the following issues for the Editor:
The title has been changed to another one that is more suited for the type of review: RELATIONSHIPS BETWEEN CHRONIC OBSTRUCTIVE PULMONARY DISEASE AND LUNG CANCER: BIOLOGICAL INSIGHTS

As answered to reviewer #1 in R7 responses, in the revised manuscript version, we would like to keep Figure 1 as it is now. In this regard, we are requesting to keep this Figure to the Editor.

English language, both style and grammar has been extensively revised throughout the entire manuscript text. We hope that the Editor and reviewers’ expectations regarding this issue will have been met in the revised manuscript version.

The legends to the Figures have also been improved in the revised manuscript version (See page 33).
Finally, it should also be mentioned that in the revised manuscript version the word count is larger (5,275 words and 124 references) than in the formerly submitted manuscript (4,162 words and 103 references) as a result of all the additional information and supporting citations that were requested by the two reviewers.

C2
Revise your manuscript using a word processing program. Please provide one clean file (no changes marked) and one marked file. You may provide this second marked file either by using the "Track Changes" function of your word processing program or by using a different color of text to show the changes.

R2
We have revised our manuscript using a Word processor. Changes to the manuscript text have been highlighted in yellow and red font has also been used for the Editors and reviewers to easily identify the modifications made.

C3
At this moment, the line-up of authorship should not be changed, according to our policy. Stealthy modification of author names will cause rejection of your paper. IF you request the need to modify the authorship with sufficient reasons, please let us know in your cover letter. AS SUCH, JOURNAL OF THORACIC DISEASE RESERVES THE RIGHT TO BEGIN THE REVIEW PROCESS ANEW.

Your kind acknowledgement of receiving this email would be greatly appreciated. We would highly appreciate if you could return the revision back within 3 weeks. If it is not possible for you to submit your revision in a reasonable amount of time, we may have to consider your paper as a new submission.

Yours sincerely,

Editorial Office

Journal of Thoracic Disease
URL:http://jtd.amegroups.com
E-mail:jtd@amepc.org

R3
We do not need to request a change to the authorship in the revised manuscript version. We are submitting the revised manuscript version within the time frame granted by the Editors. In August 8th 2016 we received the confirmation from the Editorial office (Ms Hestia) that the revised manuscript version could be submitted in August 22nd 2016, after having requested an extension of several days. We are submitting the revised manuscript today as of August 21st 2016.
LETTER TO THE REVIEWERS

Subject: Relationships Between Chronic Obstructive Pulmonary Disease And Lung Cancer

Current title in the revised manuscript version: “Relationships between chronic obstructive pulmonary disease and lung cancer: biological insights”

Reviewer A

C1
This review attempts to provide an overview on the entire Lung Cancer Field and the links between COPD and lung cancer (LC). It is a compact review and highlights in general detail some of the major biological mechanisms that may drive LC development. The strength of the review is the biological mechanisms however the beginning of the review (from page 2-10) is full of many grammatical errors, and sentences that don’t make sense which make it extremely difficult to read. The abstract also alludes to the importance of emphysema in lung cancer development but this is not clearly described or differentiated in the biological mechanism section of the review.

R1
We thank the reviewer for these interesting comments. We are also grateful to the reviewer for having considered that this review is compact and highlights the major biological mechanisms which may drive LC development in COPD. In the revised manuscript version, no differences between the clinical phenotypes emphysema versus chronic bronchitis have been made as convincing studies showing the underlying biological mechanisms are still awaited. In fact, the relationships between emphysema and lung cancer remain at the level of epidemiologic associations. Further research is still need in order to identify the underlying biology accounting for the greater predisposition of the emphysematous lung to develop lung cancer.
Moreover, we have paid attention to all the reviewer’s comments and have modified the manuscript accordingly. We have also checked the manuscript for potential stylish and grammar mistakes. The manuscript text has been revised according to both reviewers’ comments. Besides, appropriate responses to each of the reviewer’s concerns have been provided below.

Major comments:

C2
1. Grammar and sentence structure needs to be dramatically improved in the first half of the review.

R2
We thank the reviewer for this comment. We have meticulously revised the entire manuscript text and have corrected it throughout. See red font text highlighted
2. The review requires more statistical information that could strengthen the arguments of the authors such as page 6, sentence 141 beginning “Despite that” etc. What was the percentage for lung carcinogenesis versus the general population?

**R3**  
We thank the reviewer for this suggestion. Accordingly, the requested information has been provided in pages 7-8 of the revised manuscript version (under the “Radiation” subheading).

3. Page 8 sentence 181-196, this paragraph makes no sense and needs a more focused message. Conversation terms/slang such as “society nowadays” are not appropriate.

**R4**  
We thank the reviewer for this comment. The paragraph and expression have been modified in the revised manuscript version (See red font text highlighted in yellow in page 10, under “Evidence and epidemics” subheading).

4. A mention of current therapies to treat inflammation and excess oxidative stress in LC/COPD should at least be mentioned.

**R5**  
We thank the reviewer for this comment. Accordingly, information on current therapies that may target inflammation and oxidative stress in COPD and LC has been briefly provided as the focus of the present review article was not on the treatment of lung cancer, but on the links between COPD and lung cancer (See red font text highlighted in yellow in pages 12-13 and page 19).

5. The section describing Air pollution (page 7) needs greater emphasis and attention given that air pollution is a major problem in countries such as China.

**R6**  
We thank the reviewer for this comment. More emphasis and attention on air pollution has been made in the revised manuscript version (See red font text highlighted in yellow in pages 8-9, under the “Air pollution” subheading). Additionally, references related to studies conducted in China have been included in the revised manuscript version in this section (reference numbers 74-76). Moreover, it should also be mentioned that a balance has been kept between comments raised by this reviewer and those of reviewer # 2 (See C2 and R 2 responses to reviewer # 2 below), who was concerned about the extension of the manuscript, especially the first section (too extensive chapter on risk factors for LC).
C7
6. Figure 1 may not be vital. And Figure 2 and 3 require better figure legend descriptions/explanations.

R7
We thank the reviewer for this concern. We have discussed among the authors the need to keep Figure 1 in the revised manuscript version. Accordingly, we have decided to leave it as is. We believe that it will help the potential readership of the Journal to better understand the most relevant risk factors for LC. Nonetheless, we will leave the final decision to the Editor as to whether Figure q should be omitted in the revised manuscript version. Additionally, the quality of the legends to the manuscript Figures has also been improved (See modified text in page 33).

Minor comments:

C8
1. Page 1 sentence 14: delete “especially in those with emphysema”

R8
We agree with the reviewer’s suggestion and the expression has been omitted in the revised manuscript version (See red font text highlighted in yellow in page 3).

C9
2. Page 3 sentence 56: reword to “last few years by …other researcher including those listed in this review.”

R9
We thank the reviewer for this comment. Accordingly, the sentence has been modified as per requested (See red font text highlighted in yellow in page 4).

C10
3. Page 3 sentence 72: a space is required between above and mentioned.

R10
We thank the reviewer for this comment. However, it should be said that in American (US) English the word is totally correct. As the entire manuscript has been written in US English, the word has been kept the same (See red font text highlighted in yellow in page 4).

C11
4. Page 5 sentence 101: reword; sentence 107: reword; sentence 110: define cruciferous; sentence 115: reword

R11
We thank the reviewer for this comment. The paragraph under “Diet and physical activity” has been entirely revised and modified accordingly (See red font text highlighted in yellow in page 6).
5. Page 8 from sentence 176-195: Reword most of this paragraph and define the diagnostic tools.

**R12**
We thank the reviewer for this comment. The paragraph has been modified in the revised manuscript as per requested (See red font text highlighted in yellow and references provided in pages 9-10, under the “Evidence and epidemics” subheading).


**R13**
We thank the reviewer for this comment. The paragraph has been entirely modified in the revised manuscript as per requested (See red font text highlighted in yellow in page 11).

7. Page 10 sentence 245: ending, “the same patients.” More information is required. Does Th1 or Th2 increase post surgery?

**R14**
We thank the reviewer for this remark. Accordingly, the requested information has been added in this section in the revised manuscript version (See red font text highlighted in yellow in page 13, 2nd paragraph).

8. Page 11. The section reviewing Redox balance needs to comment on the previous work of others including Hodge, S et al, Bozinovski S. et al and Vlahos R et al in regards to work on oxidative stress from cigarette smoke (in COPD patients) and direct impacts on macrophages etc.

**R15**
We thank the reviewer for this comment. Accordingly, the sections on “Chronic Inflammation” and “Redox balance” have been complemented with additional references and results from other studies (as also suggested by reviewer # 2 in C3 comments below) published by renowned investigators in the field of CS, COPD, and lung cancer (See red font text highlighted in yellow in pages 12-14, 15, and 19, and references 27-29, 31, 100, 102, 111-113, 123, and 124).
Reviewer B

C1
Presented topic in the paper is novel. However following points needs attention.

R1
We are very thankful to the reviewer for the comments provided which have certainly contributed to enhancing the quality of the revised manuscript version. We have paid attention to all the reviewer’s concerns and have modified the manuscript accordingly. Moreover, responses to each of the reviewer’s comments are also provided below.

C2
1. Too extensive chapter on risk factors for lung cancer.

R2
We thank the reviewer for this comment. We also agree that this section was long. However, reviewer # 1 has asked for additional information, which has been provided in the revised manuscript version (See red font text in pages 4-9 and R6 responses provided to reviewer # 1 above). We also hope that the reviewer will agree with this additional information requested by reviewer # 1, which has been incorporated in the revised manuscript version.

C3
2. Broadly described part about redox balance and its meaning in LC progression, at relatively negligible role of the other new factors, e.g. TGF-β and its role in epithelial-mesenchymal transition, chemokine’s: CXCL5, CC21, CXCL12 and its role in migration, proliferation, angiogenesis…

R3
We thank the reviewer for this comment. Accordingly, additional information on the suggested markers and effects has been incorporated in the revised manuscript version (See red font text highlighted in yellow in page 12-19, and references 27-29, 31, 100, 102, 111-113, 123, and 124). We have included only those studies in which the suggested biological markers were analyzed in lung cancer of COPD patients or cells exposed to the effects of CS.

C4
3. Authors presented only studies concerning COPD patients with emphysema component. There are no papers with COPD with chronic bronchitis predominance, even if they are negative research.

R4
We thank the reviewer for this comment. The studies described in the “Potential biological mechanisms…” section do not necessarily refer to patients with emphysema. In fact, the referred studies were conducted in patients with COPD regardless of underlying emphysema as this was not differentiated in most of biological studies of lung cancer. As most of the investigations have been conducted in patients with COPD independently of the predominance of emphysema, modifications throughout the entire manuscript text have been carried out. In this regard, COPD irrespective of the clinical phenotypes has
been considered as the relevant risk factor for lung cancer development throughout the revised manuscript text. In specific sections, emphysema has been mentioned as the clinical phenotype that most significantly contributes to lung cancer in COPD patients, but only when referring to epidemiologic studies (See red font text highlighted in yellow in page 3, last three sentences, and page 10, 2nd paragraph). Indeed, in the revised manuscript version, no differences between the clinical phenotypes emphysema versus chronic bronchitis have been made as convincing studies showing the underlying biological mechanisms are still awaited. In fact, the relationships between emphysema and lung cancer remain at the level of epidemiologic associations. Further research is still need in order to identify the underlying biology accounting for the greater predisposition of the emphysematous lung to develop lung cancer.

C5
4. The authors have divided the manuscript into three sections. In the mind of the reader there are no difference between section two and three (review of studies), especially in abstract and introduction. It would be better to present in the second part hypothesis of the relationship between COPD and LC and support it with study evidence in third part.

R5
We agree with the reviewer’s concern. Accordingly, the objectives for the second and third sections of the review have been modified following the precise reviewer’s recommendations (See red font text highlighted in yellow in pages 2 and 4 of the Abstract and Introductions sections, respectively, in the revised manuscript version.

C6
5. The study need some stylistic corrections, e.g., “A great number of investigations has recently highlighted the relevance of emphysema in the development of LC in patients with chronic obstructive pulmonary disease (COPD), especially in those with emphysema”

R6
We thank the reviewer for this comment. Accordingly, the style and grammar have been revised and improved in the revised manuscript version (See red font text highlighted in yellow in pages 2-19). We apologize for these mistakes.

C7
6. There are some repeated fragments of text in different places of manuscript, e.g., “…the prevalence of LC can go up as high as five-fold that of smokers without the disease…”

R7
We thank the reviewer for this comment. As abovementioned the style and grammar have been revised and improved in the revised manuscript version (See red font text highlighted in yellow in pages 2-19). See modifications throughout the revised manuscript sections. We apologize for these mistakes.
Some references are old.

We thank the reviewer for this comment. An effort has been made to include recent studies (See reference list in the revised manuscript version). Nonetheless, our intention was to highlight the relevance of the problem using a few relatively old references, which support evidence of a long-lasting clinical problem that clearly still needs a solution. We have modified the paragraph in order to emphasize the actual history of the relationships between CS, COPD, and lung cancer (See red font text highlighted in yellow in pages 4-5 under the “Carcinogenic effects of CS” subheading in the revised manuscript version).

Finally, we would like to thank the reviewers again for their time and willingness to review this manuscript. All their comments have been taken into account in the revised manuscript, which has certainly been improved on the basis of the reviewers’ recommendations.
RELATIONSHIPS BETWEEN CHRONIC OBSTRUCTIVE PULMONARY DISEASE
AND LUNG CANCER: BIOLOGICAL INSIGHTS

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KEY WORDS: COPD, lung cancer, etiologic agents, epidemics, underlying biological mechanisms

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Running head: Associations between COPD and lung cancer

Word count: 5,275 words
ABSTRACT

Lung cancer (LC) has become one of the leading causes of preventable death in the last few decades. Cigarette smoking (CS) stays as the main etiologic factor of LC despite that many other causes such as occupational exposures, air pollution, asbestos, or radiation have also been implicated. Patients with chronic obstructive pulmonary disease (COPD), which also represents a major cause of morbidity and mortality in developed countries, exhibit a significantly greater risk of LC. The study of the underlying biological mechanisms that may predispose patients with chronic respiratory diseases to a higher incidence of LC has also gained much attention in the last few years. The present review has been divided into three major sections in which different aspects have been addressed: 1) relevant etiologic agents of LC, 2) studies confirming the hypothesis that COPD patients are exposed to a greater risk of developing LC, and 3) evidence on the most relevant underlying biological mechanisms that support the links between COPD and LC. Several carcinogenic agents have been described in the last decades but CS remains to be the leading etiologic agent in most geographical regions in which the incidence of LC is very high. Growing evidence has put the line forward the implications of COPD and especially of emphysema in LC development. Hence, COPD represents a major risk factor of LC in patients. Different avenues of research have demonstrated the presence of relevant biological mechanisms that may predispose COPD patients to develop LC. Importantly, the so far identified biological mechanisms offer targets for the design of specific therapeutic strategies that will further the current treatment options for patients with LC. Prospective screening studies, in which patients with COPD should be followed up for several years will help identify biomarkers that may predict the risk of LC among these patients. **Word count:** 300
INTRODUCTION

Lung cancer (LC) has become one of the leading causes of preventable death in the last few decades (1). Cigarette smoking (CS) stands as the main etiologic factor of LC despite that many other causes such as occupational exposures, air pollution, asbestos, or radiation have also been implicated (1-6). In the 1950s and 1960s, several epidemiologic studies were conducted, in which the links between CS and LC were clearly established (1;7-9). On the other hand, the combination of CS with several environmental or occupational agents may increase the risk of LC in exposed individuals (1). Presently research on the epidemiology of LC is still very active as primary prevention continues to be the most relevant target. Moreover, indoor and outdoor pollutants, which may vary with time, and the components of CS (proportions of tar and nicotine), are also a matter of research nowadays (1). Additionally, the changes in the histopathological characteristics of LC in many developed countries, with a significant rise in the frequency of adenocarcinoma, have also prompted research in this field.

Molecular epidemiology focuses on the elucidation of the biological mechanisms that favor malignancy in the lung parenchyma and airways of smokers and on the factors that enhance susceptibility to LC. In line with this, it has also been well established that patients with underlying respiratory conditions such as chronic obstructive pulmonary disease (COPD), which also represents a major cause of morbidity and mortality in developed countries, exhibit a significantly greater risk of LC (3;10-17). Importantly, in patients with moderate-to-severe COPD, the prevalence of LC can go up as high as five-fold that of smokers without the disease (18-24). Furthermore, the epidemiologic relevance of emphysema in the development of LC in patients with COPD has also been highlighted (2-5;16). The need for well-validated and practical LC screening tools to be implemented in clinical settings has also been underscored in several recent studies (2-5;17).
The study of the underlying biological mechanisms that may predispose patients with chronic respiratory diseases to a higher incidence of LC has also gained much attention in the last few years by several investigators including those listed in this review (25-31). For instance, oxidative and nitrosative stress as a result of reactive oxygen and nitrogen species (ROS and RNS, respectively) were shown to favor carcinogenesis through the activation of cellular processes that result in neoplastic transformation or the induction of DNA mutations (32). Other investigations have also demonstrated the contribution of oxidative damage, inflammatory events, and tumor microenvironment to lung carcinogenesis in patients and animal models (19;20;25-31;33-37).

The present review has been divided into three different sections in which the main topics introduced herein have been reviewed. First of all, the most relevant etiologic agents of LC are being briefly described. Secondly, the different studies that have confirmed the underlying hypothesis that COPD patients are exposed to a greater risk of developing LC are also reviewed. Finally, in the third place, evidence on the most relevant underlying biological mechanisms that support the links between COPD and LC, which account for the greater predisposition of these patients to lung tumorigenesis, is also described in the current review.

**MAIN ETIOLOGIC AGENTS OF LC**

As abovementioned, in this century, CS still continues to be the most important etiologic factor of LC. Nonetheless, several other agents such as indoor and outdoor pollutants, which may act synergistically with CS, also play a relevant role in the etiology of LC. A brief description of the most relevant etiologic factors follows below (Figure 1).

**Carcinogenic effects of CS**

We need to go back in history as far as the 1950s in order to understand the etiology of LC. Indeed, the first epidemiologic studies that demonstrated the links between the deleterious
effects of CS on the airways and lungs of smokers and LC development were published by British and American scientists in the 1950s and 1960s (1;7-9). Importantly, the duration of smoking and the number of cigarettes smoked were shown to increase the risk of LC among the smokers (38;39). Specifically, in other seminal studies, the duration of CS predicted a higher risk of LC than the amount of cigarettes smoked daily (38;39). Thus, these findings have important implications on the age at which individuals start smoking, which is presently more common in younger adolescents.

The composition of cigarettes has also changed considerably throughout time. Currently, the use of filtered cigarettes is predominant over the unfiltered cigarettes that had been used in the last century. The proportions of nicotine and tar, which includes several types of cancer promoter chemicals in the condensable residue of CS, have varied throughout time. Studies have demonstrated that in smokers consuming filtered cigarettes with lower tar proportions the risk of LC was reduced compared to those exposed to unfiltered cigarettes with higher tar yields (40-42). In another investigation (43), in smokers consuming high-yield tar cigarettes the risk of LC was significantly greater than in low- and medium-yield smokers, suggesting that tar yields are a risk factor of LC. The risks for current and past-smokers have also been reported. As such, compared to lifelong non-smokers, the risk of LC was estimated to be four-fold higher among former smokers, while it was 14-fold greater among current smokers (44). The composition of chemicals included in the cigarette may also greatly account for the pattern changes in histological types of LC that have been observed in the last few decades. In this regard, a shift towards a predominance of adenocarcinoma over squamous and small cell carcinomas has been reported since late 1970s in several investigations (45-47). Moreover, evidence has also shown that passive smokers who are exposed to second-hand smoke have a higher risk of LC, especially that of nonsmoking women with a smoker husband (48;49).
Diet and physical activity

Importantly, factors such as the diet and physical activity also seem to play a role in the risk of LC in smokers. For instance, fruits, vegetables, and antioxidant micronutrients may exert protective effects against LC (50). Results obtained from case-control and cohort studies have demonstrated that smokers with an elevated daily intake of vegetables and fruits, especially the latter, had a lower risk of LC than those subjects who did not follow this type of diet (51-53). Moreover, diets rich in other types of nutrients that are abundantly present in tomatoes and cruciferous vegetables (cross-like shape as a result of the four equal-sized petals in their flowers) such as vitamins A carotenoids, C, and folic acid, and fiber induced protective effects against LC in smokers compared to those who did not include this sort of elements in their diet (52;54-56). Despite that the studies aimed to evaluate the potential beneficial effects of certain nutrients in the reduction of LC risk in smokers are hard to conduct and interpret (confounding lifestyle factors associated with smoking), it is currently possible to conclude that vegetable consumption protects against LC (1). Finally, high levels of physical activity have also been shown to correlate with a lower risk of LC among active smokers than those with a more sedentary lifestyle, even after adjusting for CS (1). In this regard, pulmonary rehabilitation and exercise training programs for several weeks also induced beneficial effects in patients with LC who underwent thoracic surgery for the treatment of their lung neoplasm (57).

Occupational exposure

The contribution of occupational exposure to LC has been estimated to range from 9 to 15%, which is relatively low in industrialized regions when compared to the implications of CS (1). LC has been associated especially with exposure to the following agents: tar, soot, arsenic, chromium, nickel, and silica dust (1). Nonetheless, exposure to these agents has been well controlled in developed countries in the last decades (1). Additionally, in epidemiologic
studies, exposure to diesel exhaust was also demonstrated to induce LC in truck drivers (58;59), railroad workers (60), and operators of heavy construction equipment (61). More recently, exposure to new technologies included in the environment and workplace such as sandblasting jean workers and hydraulic fracturing (fracking) have introduced new hazards that may lead to the development of occupational diseases including LC (62).

Asbestos, which consists of fibers of silicate minerals, may also cause LC in exposed individuals such as coal miners (63). First evidence comes from studies conducted in the United Kingdom and United States of America, where textile (63) and insulation (64;65) workers exhibited a significantly higher risk of LC than non-exposed individuals. Importantly, asbestos-induced LC depends on the duration of the exposure and is significantly increased by the influence of CS, which may favor the retention of asbestos fibers in the lungs (66;67).

**Radiation**

Exposure to ionizing radiation has shown a strong association with LC (68). Despite that exposure to radiation produced by x-rays, gamma rays, neutrons, and radon was shown to cause LC, the levels required to induce lung carcinogenesis were significantly greater than those usually experienced by the general population (1). As such, among atomic bomb non-smoker survivors, the radiation-related risks for LC were similar to those estimated for other solid neoplasms: 0.9 with a female: male sex ratio of 1.6 (69). Radon is a chemical inert gas that occurs naturally as a decay product of radium. The decay products of radon emit alpha particles of high energy and mass that may damage nuclear DNA of cells in the lungs and airways. In fact, a very high risk of LC (40%) was observed in underground miners of uranium who had been exposed to radon chronically (70). In the study, the rates of deaths from LC were 70% and 39% for never-smokers and current smokers, respectively (70). Furthermore, in buildings, radon is ubiquitously distributed as it enters directly from the soil, and its concentrations may vary from room to room, depending on the level of ventilation.
The effects of indoor exposure to radon are significantly lower than those seen as a result of occupational exposure as in uranium miners (1;70). Recently, it has been demonstrated that residential radon may increase LC risk up to 30% among never-smokers (71;72). However, in the studies, the potential influence of environmental CS on the risk of lung cancer could not be ruled out (1;71;72). In conclusion, despite that indoor exposure to radon has been suggested to cause LC, these assumptions still need to be definitely confirmed in future epidemiologic studies (1;71;72).

**Air pollution**

The line has been recently put forward that air pollutants also contain carcinogens, which may favor LC development (1;73). Potential carcinogens include polycyclic aromatic hydrocarbons, arsenic, nickel, and chromium, which are all produced by the combustion of fossil fuels (1). Descriptive studies have highlighted a potential role of air pollution in LC development, especially in urbanized areas (1). In line with this, the relationships between long-term exposure to particulate matter < 10 micrometer in diameter (PM$_{10}$), sulfur dioxide, nitrogen dioxide and mortality of LC have been analyzed in a cohort of Northern China for several years (1998-2009) (74). The results were not conclusive as age, the assignment method for air pollution exposure, and smoking history influenced the analyses of the study results (74). In another study conducted in Tianjin (Northern China), exposure to high concentrations of polycyclic aromatic hydrocarbons induced a greater risk of LC among the elderly in a similar fashion for both men and women (75). On the other hand, ambient fine particulate matter (PM$_{2.5}$) has recently been shown to account for 32% of total reported deaths in the 74 leading cities of China (76). Specifically, 20% of the reported deaths were attributed to cardiovascular, respiratory and lung cancer conditions (76). The investigators concluded that in certain regions of China, PM$_{2.5}$ imposes significant health risks that are even greater
than those so far attributed to CS, thus action plans for Air Pollution Prevention and Control should be enforced, at least in specific geographical areas (76).

Recently, strong associations between small cell LC and adenocarcinoma (hazard ratios: 1.53 and 1.44, respectively) and exposure to high levels of PM$_{2.5}$ have also been reported in a cohort of women of the Canadian Cancer Registry (77). In other studies, however, the effects of air pollution as a risk factor of LC were significantly attenuated after adjusting for several factors such as CS and occupational exposure, even if the influence of urbanization persisted (1;78). Other approaches have included the analyses of the effects of factories and smelters in populations residing nearby. However, the results were not entirely conclusive (1). As a summary from different reports (1), it is possible to conclude that 1-2% of LC can be attributed to air pollution, although these proportions may vary widely across geographical regions.

**SUSCEPTIBILITY TO LC: INFLUENCE OF COPD AND OTHER CHRONIC RESPIRATORY CONDITIONS**

**Evidence and epidemics**

Underlying chronic respiratory conditions such as COPD and lung fibrosis, especially pneumoconiosis (1), increase the susceptibility of the patients to LC. Importantly, COPD and chronic airway obstruction have long been associated with LC development (2-5;10-13;16;79). Interestingly, the prevalence of LC e has been consistently greater in men than in females. However, in the last decades, evidence has shown a rising incidence of LC in women probably as a result of the increased numbers of female smokers, who were diagnosed with LC (1;80). Furthermore, among never-smokers the prevalence of LC in females was also significantly higher than in men (81). Most of women were diagnosed at advanced stages of
tumor progression, even among never-smokers, who exhibited similar survival rates to those reported in smokers with LC (81).

Therefore, the deployment of screening programs in patients with underlying respiratory diseases has recently emerged as an actual medical need in Western and Eastern societies (2;3;82-84). Progress has been made in the identification of the best screening programs for the early diagnosis of LC. In this regard, low-dose computerized tomography (LDCT) has been proposed as a useful screening tool for LC (85). In keeping with, several authors (3;4;86;87) have demonstrated the benefits of LDCT in the early diagnosis of LC among smokers. Nonetheless, other authors have claimed that in LC screening programs, LDCT should not be still widely applied in the general population up until more convincing results are published (2).

Interestingly, in a LC screening program using LDCT that was conducted in a region of Spain, the presence of COPD and especially of emphysema were strong predictors of LC among the study patients (3). The authors concluded that the results obtained in the LC screening program were comparable to observations that had been previously reported in other European programs, and that LDCT was a valid and feasible diagnostic tool in this context (3). In another study, the same investigators (4) showed that LC screening programs that are exclusively based on the National Lung Screening Trial (NLST) criteria may fail to identify all the cases of LC. On this basis, the investigators suggested that the application of the NLST criteria in patients with emphysema enhanced the detection rates of LC, while lowered the number of missed cases (4). Interestingly, the use of another diagnostic/therapeutic tool such as the endobronchial insertion of one-way valves for the treatment of severe dyspnea in patients with emphysema allowed for the early diagnosis of LC, especially during follow-up (82).
Risk factors of LC

With the aim to identify potential risk factors that may help predict LC morbidity and mortality other approaches have also been used in clinical settings. For instance, pleural and vascular invasion were demonstrated to influence survival and increase the risk of death in patients with non-small cell lung cancer (NSCLC) of small sizes, thus they could be used in a predictive risk model (5;84). Other observations have underscored that in patients with early stages, pneumonectomy rather than lobectomy was associated with poorer survival (12;13), and that airway obstruction was an important predictive factor to define 30-day mortality after lung surgery even in LC patients with advanced age (12;88).

Whether long-lasting effects of CS may be found in patients already diagnosed with LC has also been the matter of recent research. In this regard, in long-term survivor patients, recurrence and the appearance of second tumors were observed in the lungs and other organs of patients with LC as early as three years after the diagnosis (12;89). As the new tumors were also related to CS, the authors concluded that the multiple carcinogenic effects of CS persist several years after the LC diagnosis in patients with long survival (12;89). Thus, novel diagnostic tools are required in order to identify patients who may be at a higher risk of LC, with a special focus on patients with a long smoking history and/or the presence of underlying respiratory conditions such as COPD (90-93).

Interestingly, the risk factors of LC hospitalization have also been recently analyzed from the National Hospital Discharge Database in Spain (79). The conclusions from the study were that age and sex influenced the incidence of hospitalization in patients with LC and that while it decreased in men, a rise was observed in women, which was partly related to the presence of comorbidities (79). In keeping with, recent results obtained from the Spanish National Statistics Institute have shown that the age-adjusted mortality rates increased among Spanish women, while they decreased in men (94). These findings were related to a rise in the
prevalence of CS reported among women in Spain (94). Similar findings are probably expected in other geographical regions and deserve special attention (95;96). As a matter of fact, the incidence of CS and exposure to other carcinogenic agents represent major targets for LC prevention campaigns in Western and Eastern societies.

**POTENTIAL BIOLOGICAL MECHANISMS MEDIATING LC DEVELOPMENT IN PATIENTS WITH COPD**

**Chronic inflammation**

Chronic inflammation through the induction of several interleukins (IL) and cyclooxygenase-2 activity may be an important player in the lung tumor formation among patients with COPD (Figure 2) (97-100). Free radicals and proteases released by activated leukocytes together with the formation of tertiary lymphoid aggregates may conform the first step in tumor development of patients bearing underlying lung inflammatory conditions such as COPD (100). Moreover, these inflammatory molecules may interfere with key regulatory mechanisms such as cell death (apoptosis), autophagy, cell repair, and angiogenesis, which contribute to the neoproliferative process (97;98). Recently, migration (a key process in tumor progression) of NSCLC cells A549 was significantly increased in a chemotactic gradient produced by the serum of COPD patients compared to that of the healthy controls (29). In the serum of the patients, the concentrations of CCL21 and CXCL12, but not those of CXCL5, were significantly greater than levels found in the control subjects (29). Interestingly, the blockade of CCL21 and CXCL12 activities showed that the greater migration of the A549 cells observed in COPD was mediated by the former cytokine (CCL21) (29). The investigators concluded that CCL21 may favor cancer cell migration in the lungs of patients with COPD (29). As these results may offer an interesting therapeutic strategy to combat LC,
future studies should be specifically designed in order to demonstrate this mechanism in actual tumors from COPD patients.

Other cytokines and growth factors such as tumor necrosis factor (TNF)-alpha, vascular endothelial growth factor (VEGF), and transforming growth factor (TGF)-beta have also been shown to participate in the development of LC in patients with underlying respiratory conditions (30;101). Identification of additional inflammatory molecules that may be involved in the development of LC among patients with COPD and/or tumor progression is of relevance as they offer potential for the design of novel therapeutic strategies in the treatment of LC (100;102).

Cytokines released by T cells may play different roles in tumorigenesis. In line with this, Th1 lymphocytes, which release TNF-alpha, IL-2, and interferon-gamma, have been shown to exert antitumor effects, while Th2 cells, which mainly produce IL-4, predominantly favor tumor growth by inhibiting the host immune system (103). Importantly, LC relapse may also rely on alterations in the balance between Th1 and Th2 cytokines in patients (104-106). For instance, a rise in the systemic levels of Th2 cytokines was observed in patients with LC, while those of Th1 cytokines were decreased (106). Interestingly, after surgical resection of the lung tumor, levels of Th1 and Th2 cytokines were modified in the same patients (106). Specifically, blood levels of both IL-10 and IL-4 were significantly reduced after tumor resection compared to baseline levels before the surgery, despite that they remained significantly greater than those detected in the healthy control group of subjects (only baseline measurements) (106). A relationship between accumulation of myeloid-derived suppressor cells in patients with COPD and LC development has also been recently suggested (107). Recently, associations have also been reported between the levels of certain immune regulators in the bronchoalveolar lavage and disease progression such as metastasis and body weight loss in patients (108).
In a recent investigation from our group (unpublished observations), levels of Th1 cytokines were significantly greater in the tumors of patients with LC and underlying COPD than in those without the chronic respiratory condition. We concluded from these findings that patients with COPD may be somehow protected against tumor development and progression by the release of Th1 cytokines. Studies underway will shed light into the potential mechanisms linking the release of cytokines, chronic inflammation and the lung tumorigenesis in patients with underlying chronic respiratory conditions.

Type 1 (M1) and type 2 (M2) polarized macrophage subtypes play a significant role in tumorigenesis through the regulation of several functions such as cell adhesion, apoptosis, and senescence (104,105,109). Furthermore, macrophages may exert proinflammatory or anti-inflammatory functions depending on the secreted cytokines. In tumors, macrophages are the predominant cells within the inflammatory infiltrates. Importantly, M1 macrophages favor inflammation, whereas M2 macrophages promote anti-inflammatory actions and tissue repair. While M1 cells fight against tumor development, M2 macrophages exert the opposite effects, by promoting cancer growth, survival, progression, and dissemination (110). Recently, the profile of macrophages has been analyzed in the airways of COPD (111). As such, patients with COPD and no LC, airway macrophages did not follow the classic M1/M2 pattern, as a skewed transcriptomic profile that favors M2 macrophages was actually found (111). The authors concluded that this profile might favor tumorigenesis in COPD.

Recent unpublished observations from our group have also shown that in patients with LC, the number of M1 macrophages was reduced while a rise in M2 macrophages was observed in the same specimens. Additionally, a significantly greater M1/M2 ratio was detected in the tumors of LC patients with underlying COPD than in those of patients without this disease. These findings suggest that the prognosis of LC patients with underlying COPD
may be better than in those with no COPD. Nonetheless, further research is needed in order to confirm this hypothesis.

The implications of tumor microenvironment are also relevant in the study of the underlying biology that accounts for the greater predisposition of COPD patients to develop lung tumors. In this regard, tumor microenvironment induces immune suppression, reduces the efficacy of chemotherapy, and favors epithelial-to-mesenchymal transition (EMT) in the airways (type-II EMT, obliteration of small airways), which has recently emerged as a novel target for LC treatment (112). In line with this, a recent investigation showed that treatment of lung epithelial cells with CS extracts induced alveolar EMT through a cascade of biological mechanisms characterized by a rise in TGF-beta and Rac1/Smad2 signaling pathway (113). Interestingly, the blockade of TGF-beta activity attenuated EMT expression markers (113). The authors also concluded that these results may open a new avenue for research in the treatment of patients with LC and underlying COPD (113) as type II-EMT and angiogenesis (type-III EMT) favor lung tumor development (31).

**Redox balance**

Oxidative stress, defined as the imbalance between oxidants and antioxidants in favor of the former, represents another relevant contributing factor to LC progression (18-22;25-28;33;36) (Figure 3). Oxidative and nitrosative stress were shown to favor carcinogenesis through the activation of cellular processes that result in neoplastic transformation, the induction of DNA mutations (32;114), or even through induction of macrophage dysfunction (alterations in phagocytosis) (27). A rise in oxidant production was observed in several tissues of LC patients and in smokers (18;21;22;33). Proteins and DNA are major cellular target components for the action of oxidants that escape the cellular antioxidant systems. In this regard, several plasma proteins were strongly nitratred and oxidized in LC patients (20). Proteins involved in glycolysis, oxidant scavenging, and cellular structure were more severely
nitrated in the lung tumor tissue compared to the non-tumor parenchyma in LC patients in another study (19). More recently, patients with advanced LC exhibited increased systemic oxidative stress levels compared to healthy controls (18;33).

In a previous study from our group (25), protein carbonylation levels, as measured by malondialdehyde (MDA)-protein adducts, were also increased in the normal epithelium of patients with LC, especially in patients with underlying COPD, whose levels were significantly greater than in those without this disease. These findings were consistent with those reported in previous investigations, in which a rise in different redox markers was demonstrated in lung tissues or blood of patients with LC (19;20;25;37;101).

Several structural and functional proteins were significantly more oxidized in the lung tumors and non-tumor parenchyma in patients with LC (26). In fact, proteins such as cofilin (34), vimentin (115), and alpha-1-antitrypsin (116;117) were also shown to be more oxidized in the normal epithelium of the airways distant to the neoplasm in patients with LC (25), and their function was altered as a result of the oxidative posttranslational modifications, which may contribute to lung destruction and emphysema (116;117).

On the other hand, several proteins including vimentin, actin, and carbonic anhydrase-1 were also identified to be tyrosine nitrated in the lung tumors of patients (19). Whether these patients might also have had underlying COPD was not analyzed in that investigation (19). However, no conclusive results have been recently encountered in a study conducted in our group as protein tyrosine nitration levels did not significantly differ in the lung tumors of patients with and without COPD (19).

In a recent study (26), a significant rise in mitochondrial superoxide dismutase (SOD)2 protein content was observed in the tumors compared to non-tumor parenchyma in both groups of patients. Additionally, when patients with LC and COPD were analyzed separately on the basis of their smoking history, the heaviest smokers were those exhibiting the actual
increase in SOD2 levels in the lung tumors (26). These findings suggest that chronic CS may further enhance the rise in SOD2 content observed in the tumors of those patients. The conclusions from these findings were that SOD2 may be a key survival mechanism for the cancer cells to proliferate in the tumors. In fact, another investigation (34) demonstrated that inhibition of SOD activity reduced tumor burden in mice and promoted cell death in several NSCLC lines of cells. Furthermore, SOD2 was also shown to favor cell migration and invasiveness of tumors in other investigations (117;118). More recently, mRNA and protein levels of SOD2 were also significantly increased in lung tumors and other cancer types in patients (37). The authors concluded that SOD2 may even be considered as a biomarker for cancer progression, from tumor growth to metastasis. Hence, SOD2 overexpression seems to be involved in tumorigenesis in patients with LC, particularly in those with COPD who have a history of CS. Thus, drugs targeted to block SOD2 activity could be of interest in clinical settings.

Protein levels of SOD1 were in general much greater in the patients with underlying COPD in both tumor and non-tumor lung specimens than in LC patients with no COPD (26). Interestingly, CS did not influence SOD1 levels in any of the LC patients with COPD. The conclusions were that SOD1 seems to participate in antioxidant defense of the lungs in COPD patients regardless of the presence of LC rather than in carcinogenesis (26).

The antioxidant enzyme catalase catalyzes the decomposition of hydrogen peroxide to water and oxygen, thus protecting the cells from oxidative damage. Importantly (26), catalase protein levels were significantly reduced in the tumors compared to non-tumor parenchyma in LC patients with and without COPD. Furthermore, in LC patients with COPD, the heaviest smokers were those showing the decrease in catalase levels in the tumor lesions compared to the non-tumor lungs (26). These findings are consistent with other studies in which catalase deficiency was shown to contribute to mammary tumorigenesis in rodents (119) and cancer in...
patients (37). Collectively, it would be possible to conclude that catalase depletion seems to be involved in cancer development, especially in LC patients with underlying COPD, especially in those who were heavy smokers.

Systemic levels of the oxygen radical superoxide anion and oxidative stress markers were significantly greater, while blood levels of the antioxidant glutathione were reduced in the LC patients with COPD compared to those without this disease (26). Importantly, no significant differences between moderate and heavy smokers were seen in any of the markers analyzed in the blood samples when LC patients with COPD were further subdivided according to their smoking history. In this investigation (26), it was suggested that underlying COPD itself rather than chronic CS may account for the differential pattern of redox balance expression observed in the lung tumors and especially in the blood compartment of LC patients with and without COPD (26).

Glutathione play a relevant role in the detoxification of carcinogens and polycyclic aromatic hydrocarbons (120). Several mutations or deletions of glutathione transferases have been shown to increase susceptibility to develop cancers in patients (121;122). A meta-analysis showed that a specific genotype of glutathione transferases increased the risk to develop LC in Asian populations (120). Levels of the antioxidant reduced glutathione (GSH) were significantly lower in the plasma of LC patients with COPD than in patients without this disease. Interestingly, smoking history did not influence the results encountered in LC patients with underlying COPD, since no differences were detected between moderate and heavy smokers (26). Collectively, all these findings suggest that oxidative damage and antioxidant depletion may contribute to a greater risk to lung carcinogenesis, especially in patients with underlying COPD. Systemic levels of superoxide anion, protein carbonyls, GSH, and nitrotyrosine above a specific threshold levels were predictive of the presence of underlying COPD among the study patients with LC (26). The conclusions were that underlying COPD
may predispose patients to a higher risk to develop LC through the induction of increased levels of oxidative stress (26).

Collectively, oxidative stress appears to be a potential therapeutic target for the treatment of LC as antioxidants have recently demonstrated to exert antitumor effects by downregulating proliferating signaling pathways in experimental models (123;124). Furthermore, in the THP-1 cell line (macrophages), the antioxidant thymoquinone also attenuated the phagocytic alterations induced by CS exposure and lipopolysaccharide (27). Nonetheless, in another experimental mouse model of LC, the antioxidant N-acetyl cysteine did not show any beneficial effects on tumor growth (36). Taken together, these results suggest that more research is needed before antioxidants can be included in LC guidelines as potential therapies in actual patients.

Conclusions

Several carcinogenic agents have been described in the last decades but CS remains to be the leading etiologic agent in most of the geographical regions in which the incidence of LC is very high. Growing evidence has put the line forward the implications of COPD and especially of emphysema in LC development. Hence, COPD represents a major risk factor of LC in patients. Different avenues of research have demonstrated the presence of relevant biological mechanisms that may predispose COPD patients to develop LC. Importantly, the so far identified biological mechanisms offer targets for the design of specific therapeutic strategies that will further the current treatment options for patients with LC. Prospective screening studies, in which patients with COPD should be followed up for several years will help identify biomarkers that may predict the risk of LC among these patients.
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AUTHORS’ RESPONSIBILITY

All authors declare that the manuscript has not been submitted elsewhere, that they took a significant part in the work and have approved the final version, that they have complied with the ethical standards, and that they agree AME publishing company, to get a license to publish the accepted article when the manuscript is accepted.

AUTHORS’ CONTRIBUTIONS

EB, VB, VC, JG, JLC, XM: Conception and design

EB, JLC, XM: collection and assembly of data

EB: Manuscript writing and revised final version

EB, VB, VC, JG, JLC, XM: Intellectual input to written draft

EB, VB, VC, JG, JLC, XM: Final approval of manuscript

AUTHOR DISCLOSURE STATEMENT

The authors declare that they have no competing interests in relation to the contents of this review article.
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FIGURE LEGENDS

Figure 1: Schematic representation of the most relevant etiologic factors of lung cancer in patients. Cigarette smoking, occupational exposure (including several procedures and materials), diet, physical activity, radiation, and air pollution are the most relevant etiologic agents of lung cancer.

Figure 2: Schematic representation of the potential role of cytokines in tumor development in patients with underlying COPD. Cigarette smoking induces chronic inflammatory events characterized by the induction of several interleukins (IL), cyclooxygenase-2 activity, and cytokines. These inflammatory molecules interfere with key regulatory mechanisms such as cell death (apoptosis), cell repair, and angiogenesis, which contribute to the neoproliferative including tumor growth and metastasis.

Figure 3: Schematic representation whereby redox imbalance may induce damage in cells. Reactive oxygen species (ROS) are formed by the addition of electrons to the oxygen molecule leading to the formation of different ROS. Oxidative stress takes place in cells and tissues as a result of an imbalance between oxidants and antioxidants in favor of the former. Oxidative damage in tissues is induced through several mechanisms such as peroxidation of membrane lipids, alterations in nuclear DNA, or oxidation of cell proteins.