Respiratory tract toxicology

Exposure to chemicals by inhalation can have two effects: on the lung tissues and on distant organs that are reached after chemicals enter the body by means of inhalation. Indeed, “inhalation toxicology” refers to the route of exposure, whereas “respiratory tract toxicology” refers to target organ toxicity, in this case abnormal changes in the respiratory tract produced by airborne

Lung Structure and Function

Air enters the respiratory tract through the nasal and oral regions. The nasal passages function as a filter for particles. Highly water-soluble gases are absorbed efficiently in the nasal passages, which reach from the nostril to the pharynx. Also, nasal epithelia can metabolize foreign compounds. Cytochrome P450 isozymes 1A1, 2B1, and 4B1 have been localized in the nose in several species

Conducting Airways

The proximal airways (trachea and bronchi) of humans have a pseudostratified epithelium containing ciliated cells and two types of nonciliated cells: mucous and serous cells. Mucous cells (and glandular structures) produce respiratory tract mucus, a family of high-molecular-weight glycoproteins with a sugar content of 80% or more. They coat the epithelium with a viscoelastic sticky protective layer that traps pollutants and cell debris. Serous cells produce a fluid in which mucus may be dissolved, or upon which a mucus layer may be floated. The action of the respiratory tract cilia continuously drives the mucus layer toward the pharynx, where it is removed from the respiratory system by swallowing or expectoration. The mucus layer is also thought to have antioxidant, acid-neutralizing, and free radical scavenging functions that protect the epithelial cells.

Gas-Exchange Region

A ventilatory unit is the anatomic region that includes all alveolar ducts and alveoli distal to each bronchiolar-alveolar duct junction. Gas exchange occurs in the alveoli; adult human lungs contain an estimated 300 million alveoli. Within the alveolar septum, capillaries are organized in a single sheet. Capillaries are separated from the air space by a thin layer of tissue formed by epithelial, interstitial, and endothelial components.

Gas exchange takes place across the entire alveolar surface. A variety of abnormal processes may thicken the alveolar septum and adversely affect the diffusion of oxygen to the erythrocytes. Those processes may include collection of liquid in the alveolar space, abnormal thickening of the pulmonary epithelium, accumulation of tissue constituents in the interstitial space, and increased formation and deposition of extracellular substances such as collagen

GENERAL PRINCIPLES IN THE PATHOGENESIS OF LUNG DAMAGE CAUSED BY CHEMICALS

Toxic Inhalants, Gases, and Dosimetry

The sites of deposition of gases in the respiratory tract define the pattern of toxicity of those gases. Water solubility is the critical factor in determining how deeply a given gas penetrates into the lung. Highly soluble gases such as SO2 do not penetrate farther than the nose unless doses are very high, and are therefore relatively nontoxic . Relatively insoluble gases such as ozone and NO2 penetrate deeply into the lung and reach the smallest airways and the alveoli (centriacinar region), where they can elicit toxic responses.

Very insoluble gases such as CO and H2S efficiently pass through the respiratory tract and are taken up by the pulmonary blood supply to be distributed throughout the body.

Particle Deposition and Clearance

Particle size is usually the critical factor that determines the region of the respiratory tract in which a particle or an aerosol will be deposited. Large particles (larger than 5 μm) are usually trapped in the upper respiratory tract (nasopharyngeal region and large conducting airways), whereas smaller particles (0.2–5 μm)can be transported to the smaller airways and the alveoli .

Nanotoxicology

There is intense current interest in the lung toxicity of nanoparticles, particles with diameters of <100 nm. Ultrafine particles of this size range are increasingly being used in manufactured products, and synthesis and release of particles of this size to the environment, and exposure of individuals in the workplace, is increasing exponentially.

The toxicological concerns reflect three major issues:

1- the enormous surface area of these nanoparticles relative to their mass, especially with regard to the adsorption of co-pollutants and the presence of reactive metals on their surfaces,

2- commercially important forms of nanoparticles include nanotubes, which are high axial ratio rods that provoke concerns that they might be far more toxic than spheres of the same mass median aerodynamic diameter MMAD, and

3- the question of whether normal host defenses are effective against particles this small, which can be readily transported through and out of the lung to other tissues via pathways that are not normally accessible to larger particles.

Deposition Mechanisms

Deposition of particles occurs primarily by Interception when the trajectory of a particle brings it close enough to a surface that the particle contacts the airway surface. Interception is important for the deposition of fibers. Whereas fiber diameter determines the probability of deposition by impaction and sedimentation, interception is dependent on fiber length. Thus, a fiber with a diameter of 1 μm and a length of 200 μm will be deposited in the bronchial tree primarily by interception rather than impaction. As a result of inertia, particles suspended in air tend to continue to travel along their original path. In a bending airstream, such as at an airway bifurcation, a particle may be impacted on the surface.

At relatively symmetrical bifurcations, which typically occur in the human lung, the deposition rate is likely to be high for particles that move in the center of the airway. In the average adult, most particles larger than 10 μmin aerodynamic diameter are deposited in the nose or oral pharynx and cannot penetrate to tissues distal to the larynx. Very fine particles (0.01 μm and smaller) are also trapped relatively efficiently in the upper airways by diffusion. Particles that penetrate beyond the upper airways are available to be deposited in the bronchial region and the deep-lying airways. Therefore, the alveolar region has significant deposition efficiencies for particles smaller than 5 μm and larger than 0.01 μm.

Sedimentation brings about deposition in the smaller bronchi, the bronchioles, and the alveolar spaces. As a particle moves downward through air, buoyancy and the resistance of air act on the particle in an upward direction, while gravitational force acts on the particle in a downward direction. Eventually, the gravitational force equilibrates with the sum of the buoyancy and the air resistance, and the particle settles with a constant velocity known as the terminal settling velocity. Diffusion is important in the deposition of submicrometer particles. it is an important deposition mechanism in the nose and in other airways and alveoli for particles smaller than about 0.5 μm.

An important factor in particle deposition is the pattern of breathing. During quiet breathing, a large proportion of the inhaled particles may be exhaled. During exercise, when larger volumes are inhaled at higher velocities, deposition in airways increases. Breath holding also increases deposition. Factors that modify the diameter of the conducting airways can alter particle deposition. In patients with chronic bronchitis, the mucous layer is greatly thickened and extended peripherally and may partially block the airways in some areas. Jets formed by air flowing through such partially occluded airways have the potential to increase the deposition of particles by impaction and diffusion in the small airways. Irritant materials that produce bronchoconstriction tend to increase the tracheobronchial deposition of particles. Cigarette smoking has been shown experimentally to produce such an effect.

Nasal Clearance

Particles deposited in the nose are cleared depending on their site of deposition and solubility in mucus. Particles deposited in the anterior portion of the nose are removed by extrinsic actions such as wiping and blowing

Pulmonary Clearance There are several ways by which particulate material is removed from the lower respiratory tract once it has been deposited:

1. Particles may be directly trapped on the lining layer of the conducting

airways by impaction and cleared upward in the tracheobronchial tree via the mucociliary escalator.

2. Particles may be phagocytized by macrophages and cleared via the mucociliary escalator.

3. Particles may be phagocytized by alveolar macrophages and removed

via the lymphatic drainage.

4. Materials may dissolve from the surfaces of particles and be removed via the bloodstream or lymphatics.

5. Small particles may directly penetrate epithelial membranes.

Insoluble particles, especially long narrow fibers,may be sequestered in the lung for very long periods, often in macrophages located in the interstitium.

ACUTE RESPONSES OF THE LUNG TO INJURY
Mechanisms of Respiratory Tract Injury

The sites of interaction of toxicants in the respiratory tract have important implications for evaluation of the risk to humans posed by inhalants .Certain gases and vapors stimulate nerve endings in the nose, particularly those of the trigeminal nerve. The result is holding of the breath or changes in breathing patterns to avoid or reduce further exposure,if continued exposure cannot be avoided, many acidic or alkaline irritants produce cell necrosis and increased permeability of the alveolar walls.

Other inhaled agents can be more insidious; inhalation of HCl, NO_2, NH_3, or phosgene may at first produce very little apparent damage in the respiratory tract. The epithelial barrier in the alveolar zone, after a latency period of several hours, begins to leak, flooding the alveoli and producing a delayed pulmonary edema that is often fatal.

A different pathogenetic mechanism is typical of highly reactive molecules such as ozone. It is unlikely that ozone as such can penetrate beyond the layer of fluid covering the cells of the lung. Instead, ozone lesions are propagated by a cascade of

secondary reaction products and by reactive oxygen species that arise from free radical reactions.

Metabolism of foreign compounds can be involved in the pathogenesis of lung injury. The lung contains most of the enzymes involved in xenobiotic metabolism that have been identified in other tissues. Microsomal enzymes identified in lung include cytochrome P450 1A1, 2B1, 2F1, 4B1, and 3A4 as well as NADPH cytochrome P450 reductase, epoxide hydrolase, and flavin-containing monoxygenases. Two important cytosolic enzymes involved in lung xenobiotic metabolism are glutathione *S*-transferases and glutathione peroxidase.

Oxidative Burden

is mediated by free radicals,such as those generated by ozone, NO2, tobacco smoke, and lung defense cells , can directly and indirectly cause lung damage. Theories of lung oxidant toxicity suggest the formation of reactive and unstable free radicals and active oxygen species. Subsequent chain reactions can lead to uncontrolled destructive oxidation Recent

work has emphasized the pivotal roles of superoxide, nitric oxide ,peroxynitrate, hydroxyl radicals, & hydrogen peroxide, in mediating tissue damage. Reduction of O2 to active O2metabolites normally occurs as a by-product of cellular metabolism during both microsomal and mitochondrial electron transfer reactions; considerable amounts of superoxide anion are generated by NADPH cytochrome P-450 reductase reactions.

Mediators of Lung Toxicity

Interleukin 1 (IL-1β), transforming growth factor (TGF-beta), and tumor necrosis factor (TNF-alpha) have all been implicated in the cascade of reactions that are thought to be responsible for the pathogenesis of pulmonary fibrosis .Several members of the interleukin family, especially IL-1, IL-2, IL-5, IL-8, and IL-13, are thought to be essential components of the lung's response to epithelial cell injury. Various specific prostaglandins, especially PGE2, and leukotrienes have been implicated in intracellular signaling pathways in the lung. The roles of cell surface adhesion molecules and their interaction with cell matrix components and with control of inflammatory cell migration (particularly neutrophil influx to the lung) have been studied intensively.

Airway Reactivity

Large airways are surrounded by bronchial smooth muscles, which help maintain airway tone and diameter during expansion and contraction of the lung. Bronchial smooth muscle tone normally is regulated by the autonomic nervous system. Bronchoconstriction can be provoked by irritants such as cigarette smoke and air pollutants and by cholinergic drugs such as acetylcholine, histamine, various prostaglandins and leukotrienes, substance P, and nitric oxide. Bronchoconstriction

causes a decrease in airway diameter and a corresponding increase in resistance to airflow. Characteristic associated symptoms include wheezing, coughing, a sensation of chest tightness, and dyspnea. Exercise potentiates these problems.

Pulmonary Edema

Toxic pulmonary edema represents an acute, exudative phase of lung injury that generally produces a thickening of the alveolar-capillary barrier. Edema fluid alters ventilation-perfusion relationships and limits diffusive transfer of O2 and CO2 even in otherwise structurally normal alveoli. After exposure to some toxic chemicals in which the alveolar-capillary surface is denuded (such as alloxan), recovery is unlikely, whereas in situations of more modest injury (such as histamine administration), full recovery is readily achievable.

Accumulation and turnover of inflammatory cells and related immune responses in an edematous lung probably play a role in eliciting both mitogenic activity and fibrogenic responses.

CHRONIC RESPONSES OF THE LUNG TO INJURY
Emphysema

is commonly defined as “an abnormal enlargement of the airspaces distal to the terminal bronchiole accompanied by destruction of the walls without obvious fibrosis” these destruction results in a distended, hyperinflated lung that no longer effectively exchanges oxygen and carbon dioxide as a result of both loss of tissue and air trapping . The major cause of human emphysema is cigarette smoke inhalation, although other toxicants also can elicit this response.

A feature of toxicant-induced emphysema is severe or recurrent inflammation. The hypothesis of the pulmonary emphysema is related to alpha-1-antiprotease deficiency – which has inhibitory activity over the neutrophil elastase more promptly than over other proteinases Alpha1-antitrypsin (now called alpha1-antiprotease) is one of the body’s main defenses against uncontrolled proteolytic digestion by this class of enzymes, which includes elastase.

Studies in smokers led to the hypothesis that neutrophil (and perhaps alveolar macrophage) elastases can break down lung elastin and thus cause emphysema; these elastases usually are kept in check by alpha1-antiprotease that diffuses into the lung from the blood.

As an individual ages, an accumulation of random elastolytic events can cause the emphysematous changes in the lungs that are normally associated with aging. Toxicants that cause inflammatory cell influx and thus increase the burden of neutrophil elastase can accelerate this process.

Fibrosis

The pathological hallmark of pulmonary fibrosis is increased focal staining of collagen fibers in the alveolar interstitium. Fibrotic lungs from humans with acute or chronic pulmonary fibrosis contain increased amounts of collagen as evaluated biochemically Excess lung collagen is usually observed not only in the alveolar interstitium but also throughout the centriacinar region, including the alveolar ducts and respiratory bronchioles.

Types I and III collagen are major lung interstitial components, representing about 90% or more of the total lung collagen, that are found in the normal lungs of all mammals in an approximate ratio of 2:1. Type III collagen is more compliant than is type I; thus,increasing type I relative to type III collagen may result in a stiffer lung.

Asthma

Asthma is characterized clinically by attacks of shortness of breath, which may be mild or severe.The clinical hallmark of asthma is increased airway reactivity: the smooth muscle around the large airways contracts in response to exposure to irritants. There are well-established links between occupational and environmental exposure to antigens or to chemicals that can act as haptens and the pathogenesis of asthma air pollution (especially ultrafine particulate air pollution) as a possible cause of the observed increase in asthma.(Hapten hypothesis)

There may be common mechanisms, which are shared between asthma and pulmonary fibrosis, especially with regard to the role of recurrent or chronic inflammation in disease pathogenesis

Lung Cancer

It is now the leading cause of death from cancer among both men and women, exposure to many chemicals encountered in industrial settings pose a lung cancer risk. Inhalation of asbestos fibers and metallic dusts or fumes, such as arsenic, beryllium, cadmium, chromium, and nickel, encountered in smelting and manufacturing operations has been associated with cancer of the respiratory tract Workers who manufacture chloromethyl ether or mustard gas have an increased risk of developing lung cancers, as do workers exposed to effluent gases from coke ovens. Radon gas is a known human lung carcinogen. Formaldehyde is a probable human respiratory carcinogen. Silica, human-made fibers, and welding fumes are suspected carcinogens. Smokers who inhale radon or asbestos fibers increase their risk of developing lung cancer several fold, suggesting a synergistic interaction between the carcinogens. To what extent common air pollutants such as ozone, nitrogen dioxide, sulfur dioxide, and fumes emanating from power plants, oil refineries, and from diesel fuel powered trucks and cars contribute to the development of lung cancer in the general population remains an open question.

Damage to DNA is thought to be a key mechanism. An activated carcinogen or its metabolic product, such as alkyldiazonium ions derived from *N*-nitrosamines, may interact with DNA. However, tumors do not always develop when adducts are present, and adduct formation may be a necessary but not sufficient condition for carcinogenesis where DNA damage caused by active oxygen species is another potentially important mechanism. Ionizing radiation leads to the formation of superoxide, which is converted through the action of superoxide dismutase to hydrogen peroxide. In the presence of Fe and other transition metals, hydroxyl radicals may be formed which then cause DNA strand breaks. Cigarette smoke contains high quantities of active oxygen species and other free radicals. Additional oxidative stress may be placed on the lung tissue of smokers by the release of superoxide anions and hydrogen peroxide by activated macrophages,metabolism of carcinogens, and lipid peroxidation caused by reactive aldehydes.

AGENTS KNOWN TO PRODUCE LUNG INJURY IN HUMANS

Airborne Agents That Produce Lung Injury in Humans

Asbestos

The term asbestos describes silicate minerals in fiber form. Exposure to asbestos fibers occurs in mining operations and in the construction and shipbuilding industries. The hazards associated with asbestos exposure depend on fiber length. Fibers 2μmin length may produce asbestosis; mesothelioma is associated with fibers 5μmlong, and lung cancer with fibers larger than 10 μm. Fiber diameter is another critical feature. Fibers with diameters larger than approximately 3 μm do not readily penetrate into the peripheral lung. For the development of mesothelioma, fiber diameter must be less than 0.5 μm, because thinner fibers may be translocated from their site of deposition via the lymphatics to other organs, including the pleural surface. Once asbestos fibers have been deposited in the lung, they may become phagocytized by alveolar macrophages. Short fibers are completely ingested and subsequently removed via the mucociliary escalator. Longer fibers are incompletely ingested, and the macrophages become unable to leave the alveoli. Activated by the fibers, macrophages release mediators such as lymphokines and growth factors, which in turn attract immune-competent cells or stimulate collagen production. Asbestos-related lung disease thus may be mediated through the triggering of an inflammatory sequence of events or the production of changes that eventually lead to the initiation (DNA damage caused by reactive molecular species) or promotion (increase rate of cell turnover in the lung) of the carcinogenic process. In humans, asbestos causes three forms of lung disease: asbestosis, lung cancer, and malignant mesothelioma(a rare tumor of the cells covering the surface of the visceral and parietal pleura)

Naphthalene

Naphthalene occurs in tars and petroleum and is a widely used for synthetic tanning agents, phthalic acid anhydride, carbaryl, and 2-naphthol. It is present in ambient air. Smokers inhale substantial amounts of naphthalene in cigarette smoke.

Naphthalene epoxides may subsequently be conjugated with glutathione and form adducts that are eliminated as mercapturic acids. The epoxide can undergo rearrangement to 1-naphthol with subsequent metabolism to quinones, which are potentially toxic compounds. Naphthalene metabolites bind covalently to cellular proteins that are important in normal cellular homeostasis and protein folding and this may be related to the mechanism of toxicity by this chemical.

Blood-borne Agents That Cause Pulmonary Toxicity in Humans

Bleomycin

is a widely used cancer chemotherapeutic agent. Pulmonary fibrosis, often fatal, represents the most serious form of toxicity. The sequence of damage includes necrosis of capillary endothelial and type I alveolar cells, edema formation and hemorrhage,delayed (after 1–2 weeks) proliferation of type II epithelial cells, and eventually thickening of the alveolar walls by fibrotic changes.

In many tissues, the cytosolic enzyme bleomycin hydrolase inactivates bleomycin. In lung and skin, two target organs for bleomycin toxicity, the activity of this enzyme is low compared with that in other organs. Bleomycin stimulates the production of collagen in the lung. Before increased collagen biosynthesis, steady state levels of mRNA coding for fibronectin and procollagens are increased, presumably subsequent to a bleomycin-mediated release of cytokines such as TGF beta and TNF alpha. Bleomycin also combines with Fe (II) and molecular oxygen; when it combines with DNA, single- and double-strand breaks are produced by a free radical reaction.

Cyclophosphamide

It is an anticancer and immunosuppressive drug. The undesirable side effects include hemorrhagic cystitis and pulmonary fibrosis. Cyclophosphamide is metabolized by the cytochrome P-450 system to two highly reactive metabolites: acrolein and phosphoramide mustard. Cyclophosphamide and its metabolite acrolein initiate lipid peroxidation.

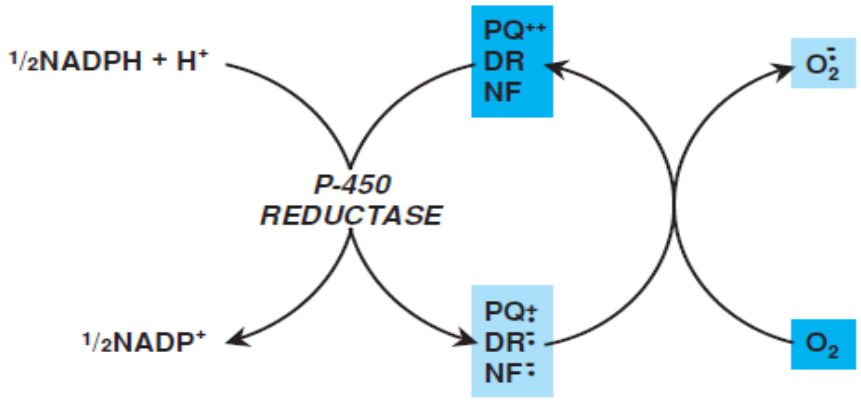

Figure 3-3. Production of superoxide anion radical ($O_2^{\bullet}$) by paraquat (PQ^{++}), doxorubicin (DR), and nitrofurantoin (NF).

Table 15-2
Industrial Toxicants That Produce Lung Disease

TOXICANT	COMMON NAME OF DISEASE	OCCUPATIONAL SOURCE	ACUTE EFFECT	CHRONIC EFFECT
Asbestos	Asbestosis	Mining, construction, shipbuilding, manufacture of asbestos-containing material		Fibrosis, pleural calcification, lung cancer, pleural mesothelioma
Aluminum dust	Aluminosis	Manufacture of aluminum products, fireworks, ceramics, paints, electrical goods, abrasives	Cough, shortness of breath	Interstitial fibrosis
Aluminum abrasives	Shaver's disease, corundum smelter's lung, bauxite lung	Manufacture of abrasives, smelting	Alveolar edema	Interstitial fibrosis, emphysema
Ammonia		Ammonia production, manufacture of fertilizers, chemical production, explosives	Upper and lower respiratory tract irritation, edema	Chronic bronchitis
Arsenic		Manufacture of pesticides, pigments, glass, alloys	Bronchitis	Lung cancer, bronchitis, laryngitis
Beryllium	Berylliosis	Ore extraction, manufacture of alloys, ceramics	Severe pulmonary edema, pneumonia	Fibrosis, progressive dyspnea, interstitial granulomatosis, lung cancer, cor pulmonale
Cadmium oxide		Welding, manufacture of electrical equipment, alloys, pigments, smelting	Cough, pneumonia	Emphysema, cor pulmonale
Carbides of tungsten, titanium, tantalum	Hard metal disease	Manufacture of cutting edges on tools	Hyperplasia and metaplasia of bronchial epithelium	Peribronchial and perivascular fibrosis
Chlorine		Manufacture of pulp and paper, plastics, chlorinated chemicals	Cough, hemoptysis, dyspnea, tracheobronchitis, bronchopneumonia	
Chromium (VI)		Production of Cr compounds, paint pigments, reduction of chromite ore	Nasal irritation, bronchitis	Lung cancer, fibrosis
Coal dust	Pneumoconiosis	Coal mining		Fibrosis
Cotton dust	Byssinosis	Manufacture of textiles	Chest tightness, wheezing, dyspnea	Reduced pulmonary function, chronic bronchitis
Hydrogen fluoride		Manufacture of chemicals, photographic film, solvents, plastics	Respiratory irritation, hemorrhagic pulmonary edema	
Iron oxides	Siderotic lung disease; silver finisher's lung, hematite miner's lung, arc welder's lung	Welding, foundry work, steel manufacture, hematite mining, jewelry making	Cough	Silver finisher's lung: subpleural and perivascular aggregations of macrophages; hematite miner's lung: diffuse fibrosislike pneumoconiosis; arc welder's lung: bronchitis
Isocyanates		Manufacture of plastics, chemical industry	Airway irritation, cough, dyspnea	Asthma, reduced pulmonary function
Kaolin	Kaolinosis	Pottery making		Fibrosis
Manganese	Manganese pneumonia	Chemical and metal industries	Acute pneumonia, often fatal	Recurrent pneumonia