Chapter 32

RICIN TOXIN

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INTRODUCTION

Ricin toxin, found in the bean of the castor plant, *Ricinis communis*, is one of the most toxic and easily produced plant toxins. It is a lectin consisting of two polypeptide chains, the A-chain and the B-chain, linked by a disulfide bond. It is one of a group of dichain ribosome-inactivating proteins, which are specific for the depurination of a single adenosine in ribosomal ribonucleic acid (RNA).[1] The active chain (ie, the A-chain) has the ability to modify catalytically the 28S subunit of eucaryotic ribosomes to block protein synthesis. The toxicity of castor beans has been known since ancient times, and more than 750 cases of intoxication in humans have been described.[2] Although ricin's lethal toxicity is approximately 1,000-fold less than that of botulinum toxin, ricin may have significance as a biological weapon because of its heat stability and worldwide availability, in massive quantities, as a by-product of castor oil production.

HISTORY AND MILITARY SIGNIFICANCE

Ricinis communis was cultivated in ancient Egypt for its oil's lubricating and laxative effects; both the oil and the whole seeds have been used in various parts of the world in the treatment of other diseases as well. During World War I and World War II, the lubricating oil was used in aircraft. Because of shortages of castor oil during World War II, the United States government subsidized the cultivation of castor beans in the San Joaquin Valley of California until the 1960s, when artificial oils replaced castor oil in the aircraft industry.

Although the industry is no longer active in this country, castor oil is still produced in large quantities throughout the world. The toxin, which remains in the castor meal after the oil has been extracted with hexane or carbon tetrachloride, is easily extracted through a simple salting-out procedure.[3]

In the late 1800s, Stillmark[4] discovered that the beans of the castor plant contained a toxic protein, which he named ricin. He discovered that ricin caused agglutination of erythrocytes and precipitation of serum proteins. (The lectin properties of ricin and abrin [a closely related toxin from the bean of *Abrus precatorius*] and their use as tools for research were described in 1972 by Sharon and Lis.[5])

Paul Ehrlich studied ricin[6] and abrin[7] during the 1890s; Ehrlich's work with these lectins became the very foundation of the discipline of immunology. Since the toxins are much less toxic when given by mouth than by injection, Ehrlich was able to induce immunity by feeding mice or rabbits small amounts of the seeds. This seminal work provided evidence that specific serum proteins are induced, capable of precipitating and neutralizing the toxin antigens. Using the same model, he also showed that, during pregnancy, specific antitoxin activity is transferred from mother to offspring through the blood and, during lactation, the same protective components are transferred through the milk.

Native ricin was first shown to inhibit tumor growth in 1951. The toxin was tested by various routes—local application, intratumor, and intra-arterial—in patients with tumors, with varying results.[8] In recent years, with the advent of new immunotherapeutic techniques, ricin has once again found a niche in the armamentarium of the medical profession. It has been studied as a component of antitumor agents called immunotoxins or, more specifically, chimeric toxins.[9]

The native ricin, or just the ricin A-chain, is conjugated to tumor cell–specific monoclonal antibodies (technically, to other ligands, which target the active component of the toxin to tumor cells for selective killing).[10] A number of these compounds have undergone Phase I or Phase II clinical trails as anticancer agents.[11,12] Although results have been promising, two factors appear to limit ricin immunotoxin efficacy: (1) lack of specificity of the antibody and (2) significant immunogenicity of the toxin moiety, which results in relatively rapid onset of refractory immunity to the therapeutic agent.[13,14]

Because of its relatively high toxicity and its extreme ease of production, ricin, code-named Compound W, was considered for weaponization by the United States during its offensive Biological Warfare Program. The U.S. Chemical Warfare Service began studying ricin as a weapon of war near the end of World War I. Work done in collaboration with the British resulted in the development of a W bomb in World War II. The weapon was tested but apparently never used in battle.[15] Ricin was used in the highly publicized assassination of Bulgarian defector Georgi Markov[16]; this incident is discussed in greater detail later in this chapter. However, because ricin intoxication is a relatively uncommon occur-

rence in human medicine, no concerted effort was made to produce specific therapies or prophylactic measures until the early 1990s, when it was perceived to be a significant biological warfare threat.

In recent years, ricin has become a favorite tool of extremist individuals or groups who seek to harm others, as the following examples demonstrate:

- Two tax protesters were convicted in February 1995 of possessing ricin as a biological weapon. This was the first case of prosecution under the 1989 Biological Weapons Anti-terrorism Act.[17]

- A retired electrician who had worked on the trans-Alaska pipeline recently committed suicide in an Arkansas jail after being arrested under the antiterrorism act for possessing castor beans. Two years before, a large quantity of ricin toxin and weapons, ammunition, and gold were found in his car by Canadian customs officials as he crossed the border from Alaska to Canada.[18,19]

Ricin's appeal to individuals such as these is likely related to its ready availability, relative ease of extraction, and its popularization by the press.[20]

DESCRIPTION OF THE AGENT

Ricin is a 66-kilodalton (kd) globular protein that makes up 1% to 5% by weight of the bean of the castor plant, *Ricinis communis*. The toxic heterodimer consists of a 32-kd A-chain that is disulfide-bonded to a 32-kd B-chain.[21] The toxin is stored in the matrix of the castor bean, together with a 120,000-d ricinus lectin.[22] Both chains are glycoproteins containing mannose carbohydrate groups; the two 32-kd chains must be associated for toxicity.

Several investigators have purified and characterized ricin[22] and have succeeded in crystallizing it. The crystal structure has been determined to 2.5 Å.[23] The A- and B-chains are globular proteins, with the A-chain tucked into a gap between two roughly spherical domains of the B-chain. A lactose disaccharide moiety is bound to each of the spherical domains of the B-chain. The disulfide bond links amino acid 259 of the A-chain and amino acid 4 of the B-chain. The crystal structure demonstrates a putative active cleft in the A-chain, which is believed to be the site of the enzymatic action of the toxin. Recombinant A- and B-chains, as well as mutants of the chains, have been expressed in *Escherichia coli* and other expression systems. The chains have been crystallized and their structures derived (Figure 32-1).[24-27]

Toxicity

There is a 100-fold variation in the lethal toxicity of ricin for various domestic and laboratory animals, per kilogram of body weight. Of animals tested, the chicken and frog are least sensitive, while the horse is the most.[28]

Toxicity of ricin also varies with route of challenge. In laboratory mice, the approximate dose that is lethal to 50% of the exposed population (LD_{50}) and time to death are, respectively, 3 to 5 µg/kg and 60 hours by inhalation, 5 µg/kg and 90 hours by
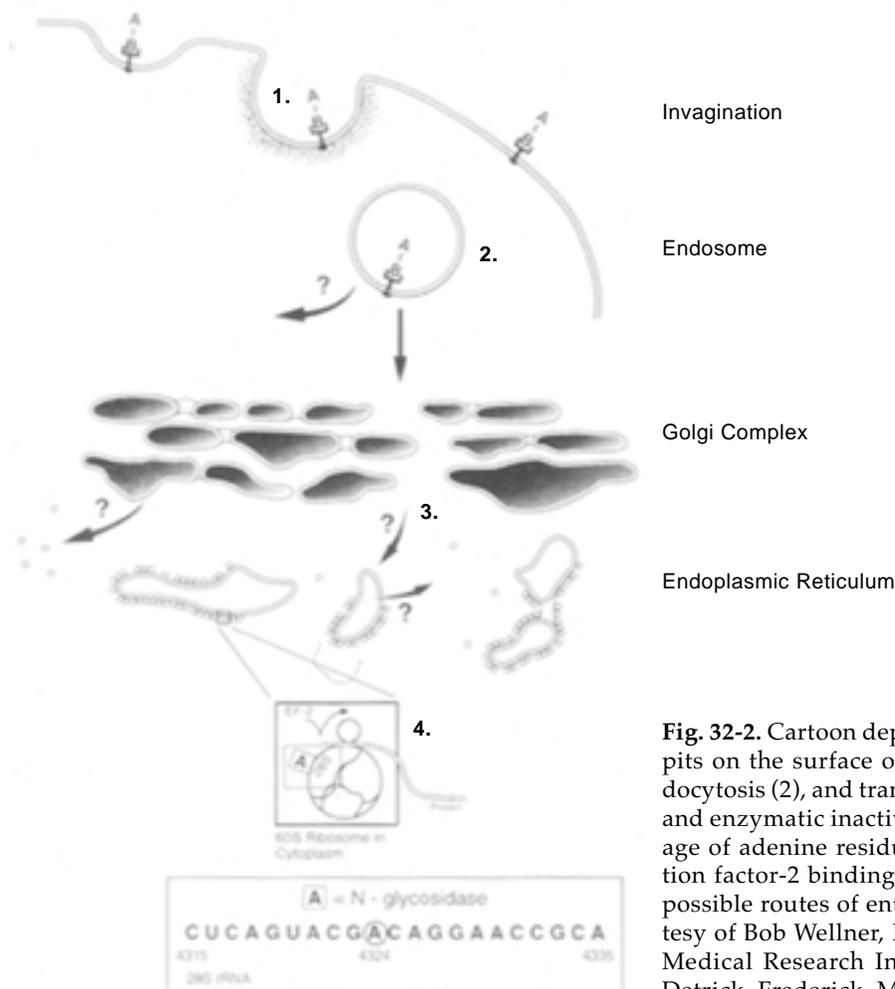

Ricin B-chain
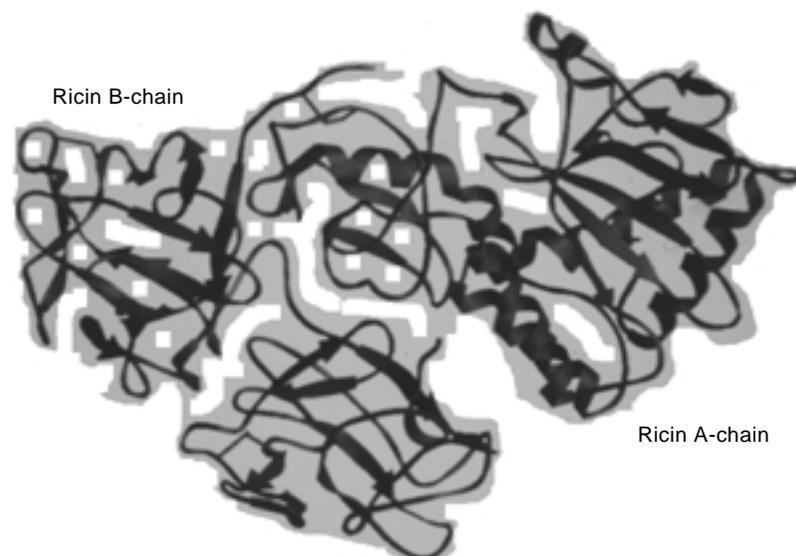
Ricin A-chain

Fig. 32-1. The crystallographically determined structure of ricin. The molecule is depicted using a ribbon representation, with the A-chain colored red; the B-chain, blue; and the disulfide bonds, yellow. Illustration: Courtesy of Mark A. Olson, PhD, Toxinology Division, US Army Medical Research Institute of Infectious Diseases, Fort Detrick, Frederick, Md.

intravenous injection, 22 µg/kg and 100 hours by intraperitoneal injection, 24 µg/kg and 100 hours by subcutaneous injection, and 20 mg/kg and 85 hours by intragastric administration. Low oral toxicity reflects poor absorption of the toxin from the gastrointestinal tract. Higher toxicities by other routes may be directly related to accessibility of target-cell populations and the ubiquity of "toxin receptors" throughout the cells of the body. When skin tests were performed on mice, no dermal toxicity was observed at the 50-µg spot.[29]

Pathogenesis

The B-chain has lectin properties that allow it to bind to complex galactosides of cell-surface carbohydrates, while the A-chain has enzymatic activity. Binding of the B-chain to glycoside residues on glycoproteins and glycolipids appears to trigger endocytotic uptake of the protein. Internalization of the toxin occurs, primarily via uncoated pits,[30,31] within a few hours; the dissociation rate of ricin for its binding sites is increased in the presence of lactose.[32] Almost all of the toxin entering the cytosol does so via the Golgi apparatus.[33] As with other protein toxins such as diphtheria toxin, *Pseudomonas* exotoxin A, and modeccin, transport to the cytosol is the rate-limiting step during the decline of protein synthesis.[34] Presence of the B-chain facilitates transport of the A-chain into the cytosol.[35] The latent periods of 1 to 3 hours in vitro and 8 to 24 hours in vivo, before cell death or clinical signs, respectively, are probably related to the necessary transport of the toxin into cells, the site of activity. Once in the cytoplasm of a eukaryotic cell, the A-chain enzymatically attacks the 28S ribosomal subunit. Ricin has a Michaelis constant (K_M) of 0.1 µmol/L for ribosomes and an enzymatic constant (K_{cat}) of 1,500/min. Ricin cleaves one adenine residue (A4324) near the 3´ end of 28S RNA. This deletion causes elongation factor-2 to fail to bind and thereby blocks protein synthesis.[36,37] At the cellular level, ricin kills through inhibition of protein synthesis (Figure 32-2).

1.   Invagination

2.   Endosome

3.   Golgi Complex

       Endoplasmic Reticulum

4.

A = N - glycosidase

CUCAGUACG(A)CAGGAACCGCA

Fig. 32-2. Cartoon depicting ricin (A—B) binding to coated pits on the surface of the cell (1), internalization via endocytosis (2), and transport through the Golgi complex (3); and enzymatic inactivation of protein synthesis via cleavage of adenine residue (A4324), and blockage of elongation factor-2 binding (4). Arrows with "?" indicate other possible routes of entry to the cytosol. Illustration: Courtesy of Bob Wellner, PhD, Toxinology Division, US Army Medical Research Institute of Infectious Diseases, Fort Detrick, Frederick, Md.

CLINICAL SYMPTOMS, SIGNS, AND PATHOLOGY

The clinical signs, symptoms, and pathological manifestations of ricin toxicity vary with the dose and the route of exposure.[38] Experimental animal studies indicate that clinical signs and pathological changes are largely route specific; for example, inhalation results in respiratory distress and airway and pulmonary lesions; ingestion causes gastrointestinal signs and gastrointestinal hemorrhage with necrosis of liver, spleen, and kidneys; and intramuscular intoxication causes severe localized pain, muscle and regional lymph node necrosis, and moderate involvement of visceral organs. The route-specific pathology is probably due to the lectin properties of ricin, which cause it to bind rapidly to complex galactosides of cell-surface carbohydrates. Transient leukocytosis appears to be a constant feature in humans, whether intoxication is via injection or oral ingestion. Leukocyte counts 2- to 5-fold higher than the normal value are characteristic findings in cancer patients, and also occurred in the Markov case.[16]

Oral Intoxication

Ricin is less toxic by oral ingestion than by other routes, probably because of poor absorption and some enzymatic digestion in the digestive tract. In oral (and parenteral) intoxication, cells in the reticuloendothelial system, such as Kupffer cells and macrophages, are particularly susceptible, due to the mannose receptor present exclusively in macrophages.[39] In 1985, A. Rauber and J. Heard[2] summarized the findings from their study of 751 cases of castor bean ingestion. There were 14 fatalities in this study, constituting a death rate of 1.9%—much lower than traditionally believed. Twelve of the 14 cases resulting in death occurred before 1930. Even with little or no effective supportive care, the death rate in symptomatic patients has been low—in the range of 6%. The reported number of beans taken by patients who died varied greatly. Of the two cases of lethal oral intoxication documented since 1930, one was of a 24-year-old man who ate 15 to 20 beans, and the other was a 15-year-old boy who ate 10 to 12 beans. All of the reported serious or fatal cases of castor bean ingestion have the same general clinical history: rapid (less than a few hours) onset of nausea, vomiting, and abdominal pain; followed by diarrhea; hemorrhage from the anus; anuria; cramps; dilation of the pupils; fever; thirst; sore throat; headache; vascular collapse; and shock. Death occurred on the third day or later. The most common autopsy findings in oral intoxication are multifocal ulcerations and hemorrhages of gastric and small-intestinal mucosa, which may be quite severe; lymphoid necrosis in the mesenteric lymph nodes, gut-associated lymphoid tissue (GALT), and spleen; Kupffer cell and liver necrosis; diffuse nephritis; and diffuse splenitis.

Injection

In one large clinical trial,[40] low doses (18–20 µg/m²) of intravenous ricin administered to cancer patients were well tolerated. Flulike symptoms with fatigue—in some cases very pronounced fatigue—and muscular pain were common, and sometimes nausea and vomiting occurred. The symptoms began 4 to 6 hours after administration and lasted for 1 to 2 days. Two toxic deaths were reported in Phase I clinical trails of the closely related protein toxin, abrin; these patients had general seizures and other signs of central nervous system toxicity.

In the case of Mr. Markov, whose assassination was mentioned earlier,[16] injection of a lethal dose of ricin, estimated to be as much as 500 µg, resulted in almost immediate local pain, then a feeling of weakness within about 5 hours. Fifteen to 24 hours later, he had a high temperature, nausea, and vomiting. Thirty-six hours after the incident, he was admitted to the hospital feeling very ill. He had fever, tachycardia, and normal blood pressure; lymph nodes in the affected groin were swollen and sore; and a 6-cm diameter area of induration and inflammation was observed at the injection site on his thigh. Just over 2 days after the attack, he became suddenly hypotensive and tachycardic; the pulse rate was 160 beats per minute, and vascular collapse and shock had set in. His white blood count was 26,300/mm³. Early on the third day after the attack, he became anuric and began vomiting blood. An electrocardiogram demonstrated complete atrioventricular conduction block. Mr. Markov died shortly thereafter; at the time of death, his white blood count was 33,200/mm³.

Intramuscular or subcutaneous injection of high doses of the toxin in humans, as occurred in the assassination, results in severe local lymphoid necrosis, gastrointestinal hemorrhage, liver necrosis, diffuse nephritis, and diffuse splenitis. In the case of Mr. Markov, a mild pulmonary edema was thought to have been secondary to cardiac failure. Similar results have been reported in experimental animal studies.

Inhalation

Although data on aerosol toxicity exposure are not available for humans, lesions induced by oral and parenteral exposure are consistent with those seen in experimental animal studies, suggesting that the same would hold true for aerosol exposures. The only information on inhalation of ricin in humans is an allergic syndrome reported in workers exposed to castor bean dust in or around castor oil processing plants.[41] The clinical picture is characterized by sudden onset of congestion of the nose and throat, itchiness of the eyes, urticaria, and tightness of the chest. In more severe cases, wheezing, leading to bronchial asthma, may also occur, and may last for several hours. Affected individuals respond to symptomatic therapy and removal from the source of exposure.

Inhalational exposure of rats to lethal ricin challenge results in a diffuse necrotizing pneumonia of the airways, with interstitial and alveolar inflammation and edema.[42] No notable changes in lung injury parameters occur before 8 hours after the challenge. By 12 hours, inflammatory cell counts and total protein (both from fluid obtained via bronchoalveolar lavage) increase, suggesting both increased permeability of the air–blood barrier and cytotoxicity; these findings are associated with a blood-cell analysis indicative of inflammation. By 18 hours after the challenge, alveolar flooding is present, and extravascular lung water is increased. Both continue to increase up to 30 hours after the challenge. At 30 hours after the challenge, arterial hypoxemia and acidosis are present and histopatho-

logical evidence of alveolar flooding becomes significant. Recently completed immunohistochemical studies[43] in rats exposed to ricin via aerosol indicate that aerosolized ricin binds to ciliated bronchiolar lining cells, alveolar macrophages, and alveolar lining cells (Figure 32-3).

In a recent study[44] of nonhuman primates, inhalational toxicity was characterized by a dose-dependent preclinical period of 8 to 24 hours, followed by anorexia and progressive decrease in physical activity. Death occurred 36 to 48 hours after the challenge, time to death also being dose dependent. Relevant gross and histopathological changes were confined to the thoracic cavity (Figure 32-4). All monkeys had acute marked-to-severe fibrinopurulent pneumonia, with variable degrees of diffuse necrosis and acute inflammation of airways. There were also diffuse, severe alveolar flooding and peribronchovascular edema (Figure 32-5), acute tracheitis, and marked-to-severe purulent mediastinal lymphadenitis. Two monkeys had acute adrenalitis.

Cause of Death

The exact cause of death is unknown and probably varies with route of intoxication. Results of ricin A-chain immunotoxin clinical trial studies demonstrated that ricin A-chain caused a vascular leak syndrome characterized by hypoalbuminemia and edema; subsequent in vitro studies with human umbilical vein endothelial cells showed that ricin damaged endothelial cells.[45] In mice, rats, and primates, high doses via inhalation appear to produce severe enough pulmonary damage to cause death,
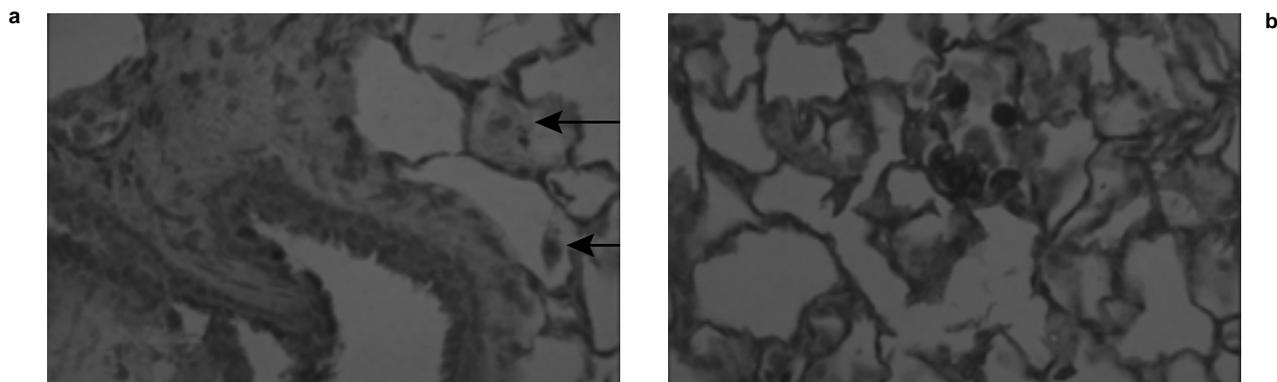
Fig. 32-3. Lung from a rat exposed to ricin by aerosol. Immunocytochemical stain for ricin demonstrates strong reactivity for (**a**) airway epithelial cells and alveolar macrophages (arrows) and (**b**) alveolar lining cells. Original magnification x 50, immunocytochemical stain. Photographs: Courtesy of CL Wilhelmsen, DVM, PhD, Lieutenant Colonel, Veterinary Corps, US Army; Division of Pathology, US Army Medical Research Institute of Infectious Diseases, Fort Detrick, Frederick, Md.
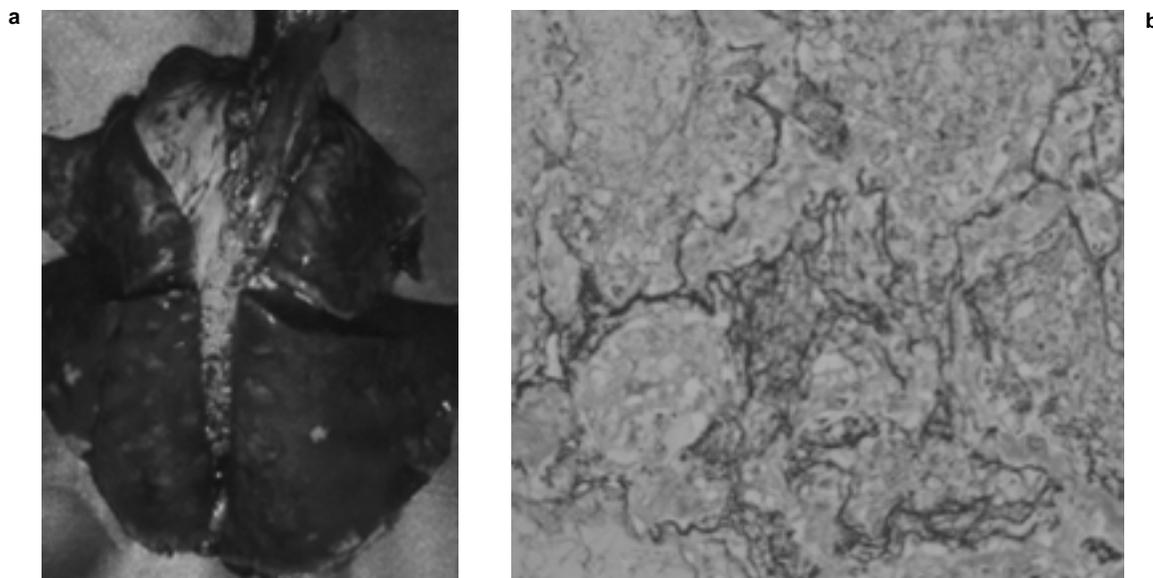
Fig. 32-4. Lungs from a monkey exposed to ricin by aerosol. (**a**) The lungs are edematous, with accompanying hemorrhage and necrosis. (**b**) Histologically, the microscopical changes are characterized by fibrinopurulent pneumonia. The fibrin has been specifically stained by phosphotungstic acid hematoxylin to appear purple (original magnification x 25). Photographs: Courtesy of CL Wilhelmsen, DVM, PhD, Lieutenant Colonel, Veterinary Corps, US Army; Division of Pathology, US Army Medical Research Institute of Infectious Diseases, Fort Detrick, Frederick, Md.
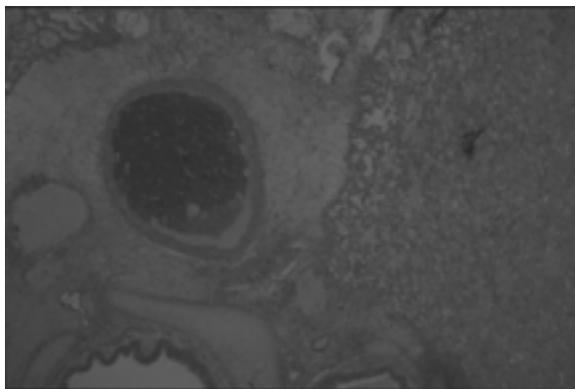
Fig. 32-5. Widespread perivascular and peribronchiolar edema in a monkey, a characteristic finding in aerosol ricin intoxication (hematoxylin-eosin stain; original magnification x 10). Photograph: Courtesy of CL Wilhelmsen, DVM, PhD, Lieutenant Colonel, Veterinary Corps, US Army; Division of Pathology, US Army Medical Research Institute of Infectious Diseases, Fort Detrick, Frederick, Md.

probably due to hypoxemia resulting from massive pulmonary edema and alveolar flooding. High doses administered intravenously in experimental animals are associated with disseminated intravascular coagulation[8]; it has been suggested that hepatocellular and renal lesions are the result of vascular disturbances induced by the toxin rather than a direct effect of the toxin itself.[46] Studies published in 1987[47] clearly establish that in intravenous administration of ricin toxin to rats, diffuse Kupffer cell damage occurred within 4 hours, followed by endothelial cell damage, formation of thrombi in the liver vasculature, and finally, hepatocellular necrosis, confirming studies that had been published in 1976.[48]

DIAGNOSIS

Like other potential intoxications on the unconventional battlefield, epidemiological findings will likely play a central role in diagnosis. The observation of multiple cases of very severe pulmonary distress in a population of previously healthy young soldiers, linked with a history of their having been

at the same place and time during climatic conditions suitable for biological warfare attack, would be suggestive.

The differential diagnoses of aerosol exposure to ricin would include staphylococcal enterotoxin B, exposure to pyrolysis by-products of organofluorine polymers (eg, Teflon [polytetrafluoroethylene, manufactured by Du Pont Polymers, Wilmington, Delaware], Kevlar [polyparaphenyleneterephthalamide, manufactured by Du Pont Advanced Fiber Systems, Wilmington, Delaware]) or other organohalides, oxides of nitrogen, and phosgene. Insecticides such as paraquat and α-naphthylthiourea (ANTU), although not expected in a battlefield scenario, can be spread aerially over large geographical areas and are also potent edemagenic agents.

Confirmation of ricin inhalational intoxication would most likely be through enzyme-linked immunosorbent assay analysis of a swab sample from the nasal mucosa; ricin can be identified by this method for at least 24 hours after the challenge.[49] Because ricin is extremely immunogenic, individuals surviving a ricin attack would likely have circulating antibody within 2 weeks after the exposure; serum samples should be obtained from survivors. Following inhalational intoxication in laboratory animals, laboratory findings are generally nonspecific.

Enzyme-linked immunosorbent assays (for blood or other body fluids)[50] or immunohistochemical techniques (for direct analysis of tissues) may be useful in confirming ricin intoxication. However, because ricin is bound very quickly regardless of route of challenge, and metabolized before excretion, identification in body fluids or tissues is difficult. In rats exposed to ricin labeled with iodine 125 by intravenous injection, the radioactive label was found in liver (46%), muscle (13%), and spleen (9%) 30 minutes after intravenous injection.[51] Ricin was quickly cleared from the animals, with only 11% remaining after 24 hours; 70% was excreted in the urine as low-molecular-weight metabolites. Attempts at identification of the toxin may also include introduction of biological autopsy materials into mice or cultured cells and neutralization through the use of specific antibodies.

MEDICAL MANAGEMENT

The most likely scenarios in which ricin intoxication might be seen by military medical personnel are (1) small-scale battlefield or terrorist delivery of an aerosol and (2) parenteral administration of the toxin to an individual as an assassin's tool. Because ricin acts rapidly and irreversibly (directly on lung parenchyma after inhalation, or is distributed quickly to vital organs after parenteral exposure), postexposure therapy is more difficult than with slowly processed, peripherally acting agents (such as the botulinum toxins or bacterial agents) that can be treated with antibiotics. Therefore, immunization of personnel at risk for ricin exposure is even more important than it is for some of the other potential biological warfare agents.

Immunization and Passive Protection

Animal studies have shown that either active immunization or passive prophylaxis or therapy (if the therapy is given within a few hours) is extremely effective against intravenous or intraperitoneal intoxication with ricin. On the other hand, inhalational exposure is best countered with active immunization or prophylactic administration of aerosolized specific antiricin antibody. Active prophylaxis through immunization is the only effective medical countermeasure within our grasp at this time.

Prophylactic immunization of mice, rats, and subhuman primates with two to three doses of the toxoid (3–5 μg per dose), with or without adjuvant (aluminum hydroxide), protects against death and incapacitation following inhalational exposure to multiple lethal doses of ricin toxin.[52] Either a toxoid of the native toxin or a preparation of the purified A-chain produces a measurable antibody response that correlates with protection from lethal aerosol exposure. The toxoid has been microencapsulated in glactide-glycolyde microparticles and, in mice, provides effective immune protection after one immunization.[53] As this is written, the only potential approved medical countermeasures for human use are the formalin-treated toxoid, which has gone through preclinical testing and has been submitted to the Food and Drug Administration as an Investigational New Drug, and the deglycosylated A-chain, which has also shown promise as an antigen in preclinical trials. The toxoid has proven safe and effective in rodents and nonhuman primates exposed to ricin aerosol by inhalation, the route of challenge believed to be the most likely threat on the battlefield.

In seeking an approach to passive protection of soldiers without immunization, animal studies have been conducted in the laboratory to evaluate the short-term efficacy of prophylactically inhaled spe-

cific antibody. Preliminary data[54] suggest that (*a*) when mice are exposed for 20 minutes to aerosolized specific immunoglobulin G (about 24 µg antibody per mouse, the lowest dose tested), they are completely protected from lethal aerosol challenge 1 hour later; and (*b*) more than 95% of them are protected from pulmonary lesions. These findings suggest that inhalation of protective antibody from a portable nebulizer just before an attack might provide some protection in unimmunized individuals. These studies also suggest that intravenous administration or inhalation of specific antibody after exposure to aerosolized ricin will be of little value in blocking or reversing the toxin's pathological effects.

Supportive and Specific Chemotherapy

As is the case in toxicity and pathogenesis of intoxication, the route of exposure is important in relation to possible modes and their likelihood of success of prophylaxis and therapy. For oral intoxication, supportive therapy includes activated charcoal administration and intravenous fluid and electrolyte replacement. For inhalational intoxication, supportive therapy to counteract acute pulmonary edema and respiratory distress is indicated. Symptomatic care is the only intervention presently available to clinicians for the treatment of incapacitating or lethal doses of inhaled ricin. Positive end-expiratory ventilatory therapy, fluid and electrolyte replacement, antiinflammatory agents, and analgesics

would likely be of benefit in treating aerosol-intoxicated humans. As we learn more about the pathogenesis of intoxication by this route, specific mediator blocking agents may prove valuable, as well.

In recent years, a wide variety of chemotheraputic agents has systematically been screened in an in vitro Vero cell inhibition of protein synthesis assay for efficacy against ricin toxicity.[55] More than 150 agents, including cellular membrane effectors, calcium channel–blocking agents, sodium–calcium exchangers, reducing agents, antioxidants, effectors of endocytosis, nucleoside derivatives, antibacterials, ricin analogs, effectors of cellular metabolism, and competitors for binding have been tested. Of these, only five agents showed promise in vitro and were screened in mouse-protection assays:

- D-galactose and its derivatives, which are competitors for binding,
- azidothymidine (AZT) and a purine derivative, BM33203, both of which are nucleoside derivatives, and
- brefeldin-A, which is a Golgi transport inhibitor.

None of the compounds have proven useful for protecting laboratory animals, even from intravenous exposure to the toxin. Efforts are also underway to synthesize very specific compounds, transition-state inhibitors, which block the enzymatic effects of the A-chain.

SUMMARY

Ricin is a large, moderately toxic, protein di-chain toxin from the bean of the castor plant, *Ricinis communis*. It can be produced easily in relatively large quantities. Ricin was developed as a biological weapon by the United States and its allies during World War II. Although ricin is toxic by several routes, when inhaled as a respirable aerosol, it causes severe necrosis of the airways and increased permeability of the alveolar-capillary membrane. The inhalational route

is presumed to be the likeliest threat on the battlefield.

Death after inhalation of a lethal dose appears to be caused by hypoxemia resulting from massive pulmonary edema and alveolar flooding. Diagnosis can be confirmed through the use of enzyme-linked immunosorbent assays of tissues or body fluids. Prophylactic administration of an investigational vaccine protects laboratory animals from inhalational and other routes of challenge.

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