Ricin and *Ricinus communis* in pharmacology and toxicology—from ancient use and “Papyrus Ebers” to modern perspectives and “poisonous plant of the year 2018”

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Received: 18 April 2019 / Accepted: 4 July 2019 / Published online: 29 July 2019
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Abstract

While probably originating from Africa, the plant *Ricinus communis* is found nowadays around the world, grown for industrial use as a source of castor oil production, wildly sprouting in many regions, or used as ornamental plant. As regards its pharmacological utility, a variety of medical purposes of selected parts of the plant, e.g., as a laxative, an anti-infective, or an anti-inflammatory drug, have been described already in the sixteenth century BC in the famous Papyrus Ebers (treasured in the Library of the University of Leipzig). Quite in contrast, on the toxicological side, the native plant has become the “poisonous plant 2018” in Germany. As of today, a number of isolated components of the plant/seeds have been characterized, including, e.g., castor oil, ricin, *Ricinus communis* agglutinin, ricinin, nudiflorin, and several allergenic compounds. This review mainly focuses on the most toxic protein, ricin D, classified as a type 2 ribosome-inactivating protein (RIP2). Ricin is one of the most potent and lethal substances known. It has been considered as an important bioweapon (categorized as a Category B agent (second-highest priority)) and an attractive agent for bioterroristic activities. On the other hand, ricin presents great potential, e.g., as an anti-cancer agent or in cell-based research, and is even explored in the context of nanoparticle formulations in tumor therapy. This review provides a comprehensive overview of the pharmacology and toxicology-related body of knowledge on ricin. Toxicokinetic/toxicodynamic aspects of ricin poisoning and possibilities for analytical detection and therapeutic use are summarized as well.

Keywords *Ricinus communis* · Papyrus Ebers · Ricin · Castor oil · Intoxication · Nanoparticles · Immunotoxins · Tumor therapy

Abbreviations

BC	Before Christ
BTWC	Biological and Toxin Weapons Convention
CB-1A	Castor bean allergenic fraction
CDC	Centers for Disease Control and Prevention

Electronic supplementary material The online version of this article (<https://doi.org/10.1007/s00210-019-01691-6>) contains supplementary material, which is available to authorized users.

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CEA	Carcinoembryonic antigen
CNS	Central nervous system
CWC	Chemical Weapons Convention
ELISA	Enzyme-linked immunosorbent assay
EMA	European Medicines Agency
EPR	Enhanced permeability and retention
ER	Endoplasmic reticulum
EROEI	Energy return-on-energy investment
EQuATox	Establishment of Quality Assurance for the Detection of Biological Toxins of Potential Bioterrorism Risk
FAO	Joint Food and Agriculture Organization
FDA	Food and Drug Administration
H5WYG	Histidine-rich fusogenic peptide
HBV	Hepatitis B virus
HER2/neu	Human epidermal growth factor receptor 2
IgG	Immunoglobulin G
i.m.	Intramuscular
i.v.	Intravenous

IUPAC	International Union of Pure and Applied Chemistry
LD ₅₀	Lethal dose 50%
LFA	Lateral flow assay
LC/MS	Liquid chromatography/mass spectrometry
MALDI-TOF	Matrix-assisted laser desorption/ionization time-of-flight
MR	Mannose receptor
MWNTs	Multiwalled carbon nanotubes
OPCW	Organization for the Prohibition of Chemical Weapons
PAP	Pokeweed anti-viral proteins
PCR	Polymerase chain reaction
PEG	Polyethylene glycol
ROS	Reactive oxygen species
RCA	<i>Ricinus communis</i> agglutinin
RIP2	Ribosome-inactivating protein type 2
RTA	Ricin A chain
RTB	Ricin B chain
RNAi	RNA interference
VLPs	Virus-like particles
VLS	Vascular lead syndrome
WHO	World Health Organization

Introduction

The native plant *Ricinus communis* probably originates from Africa (Ethiopia) or India and is considered as an ancient oil plant of Egypt. Nowadays being distributed worldwide, it is also known under other names, e.g., “Palma Christi” or “wonder tree.” It has a very long and interesting history. The name wonder tree is found in the biblical book Jonah because of the plant’s rapid growth and in the description of the ricinus-feeding worm (Jonah 4; book Jona, chapter 4):

So Jonah went out of the city [Nineveh] ... And the Lord God appointed a castor oil plant [Hebrew: kikayon], and made it to come up over Jonah, that it might be a shade over his head, to deliver him from his distress. And Jonah was exceeding glad of the plant. But God appointed a worm [Hebrew: tola'at] when dawn came up the next day, and it attacked [literally: beat] the plant, so that it withered. And it came to pass, when the sun arose, that God prepared a vehement east wind, and the sun beat down upon the head of Jonah, so that he fainted. (from Hausmann and Müller (2006); see references therein).

The name “Palma Christi” or “Palm of Christ” is based on its ability to heal wounds and cure ailments (Roxas-Duncan and Smith 2012). Indeed, different components of the plant have

been described as sanatory ingredients already more than 3500 years ago in the famous Papyrus Ebers which is the only completely preserved papyrus scroll on the ancient Egyptian healing arts in hieratic scripture (Scholl 2002). Interestingly, in Egypt, seeds have been described as ancient means of payment. Already in the 4th millennium BC, ricinus seeds were utilized as grave goods (Germer 1982), indicating their importance for the deceased and the life after death.

The Latin name *ricin* was first mentioned by the natural scientist Plinius the Elder in the first century in his opus on natural history (NH 15.7.25). It corresponds to the Greek name κροτών, meaning “tick” (Latin: *Ixodes ricinus*), as well as the ricin tree, and when used in papyri in the plural form, the seeds. According to Plinius, the tree’s name is based on the resemblance of its seeds with ticks. The artepitheton “communis” may indicate its broad distribution (“common”). In the German region, it was first mentioned by Albertus Magnus († 1280 in Cologne), who also cultivated the plant (“arbor mirabilis”). A first botanical description of the plant and its medical characteristics were given by Hieronymus Bock (1498–1554) (von Fischer-Benzon 1894). According to its use as a rich source of plant oil, the plant and the seeds are also named “castor (oil) plant” and “castor bean”, respectively, with the latter also indicating that it resembles a bean (Fabaceae) (Buch 2018 and references therein).

Nowadays, *Ricinus communis* is found not only in tropical and subtropical regions but also as a wild sprouting or ornamental plant essentially around the world. As a source for castor oil production, the plant is mentioned in connection with different compositions for cosmetic and medical products or within various applications in the industry. Parts of the plant as well as the castor oil itself are among the oldest drugs and have been used in traditional or folk remedies for rituals of sacrifice. Among indigenous peoples (e.g., in Nepal), the use of ricinus seeds has been described as an oral contraceptive (women eat 1–2 beans daily during their fertile days). In alternative medicine, the blow-promoting effect of castor oil is known and explored as so-called blow cocktail in obstetrics. There are also descriptions on its use as a human nutritional supplement, an emetic, or a purgative as well as for treating a wide range of diseases around the world (e.g., Scarpa and Guerci 1982; Poelchen and Wirkner 2003; Olsnes 2004; Rana et al. 2012; Roxas-Duncan and Smith 2012; Tunaru et al. 2012; Abdul et al. 2018; <https://www.naturinstitut.info/rizinusoel.html>).

On the other hand, the highly toxic component ricin can be isolated from castor beans as well, which is one of the most effective plant toxins (for comparison, see Suppl. Table 1 for examples of representative LD₅₀ values of different kinds of toxins). In fact, for a long time, it has been explored as a possible biological weapon and is still relevant as a bioterrorist agent.

According to Fritz Hauschild, ricin is one of the five most toxic substances known: tetanus toxin, botulinus toxin,

diphtheria toxin, gramicidin, and ricin (Hauschild 1960). It may thus not come as a surprise that *Ricinus communis* has been awarded the title “poisonous plant 2018” in Germany—providing the ideal occasion for a critical appraisal of its relevance (“[Giftpflanze des Jahres 2018](https://www.hamburg.de/wandsbek/giftpflanze-des-jahres)”; Botanischer Sondergarten Wandsbek: <https://www.hamburg.de/wandsbek/giftpflanze-des-jahres>).

History-Papyrus Ebers and the plant *Ricinus communis*

The text from the Papyrus Ebers, the longest and completely preserved papyrus role dealing with the ancient Egyptian art of healing and treasured in the Library of the University of Leipzig, is an informative and unique description dated from the last quarter of the sixteenth century BC. Indeed, it indicates the existence of textual descriptions on the medical use of plants already in the Pharaonic era. The text passages dealing with the ricinus plant (Eb 251, column 47, line 15–column 48, line 3) are often named as “ricinus book” and summarize its curative applications (Fig. 1, column 47, line 15–22). Thus, rather than describing the phenotype of the plant, it focuses on an enumeration of which parts and products of it are useful for treating various maladies. The ancient Egyptian word for ricinus is (in transcription) “dgm”. In contrast to older literature that had identified the plant “k3k3” as ricinus, more recent research, in particular by the Saxonian Academy of Sciences, indicates “dgm” as the correct word, with “k3k3” rather meaning “bushes” in general. Several parts of the plant (radix, seeds, castor oil) have been described in detail for a variety of medical purposes.

Eb 251 (47, 15 - 48, 3): Knowledge of what is made out of the ricinus plant (dgm), as something found in the writings of old, as something useful to man: a) One crushes its roots in water; they are applied to a sick head; he (the patient) gets immediately well like someone who is not ill; b) a little of its fruit is also chewed with beer by one with the whj-symptom, (diarrhoea, Med. Gram. § 299 & G. IX, 72) in his stools; this is an elimination of suffering (b-j-t) in man's belly; c) the growth of woman's hair is also promoted by its seed; it is ground, made into a mass and added to oil (mr/:1:t); then the woman should anoint (gs) her head with it; d) its oil (mr/:1:t) is also prepared from its seed to anoint (gs) (a man) who (has) the w/:1}w-skin disease (and) is affected with i!N (and) bw~w, being unwell.¹

¹ Here and in the following, the English translations of the Papyrus Ebers are taken from Ghalioungui (1987).

The credibility of this knowledge is supported by the indication of its existence already in ancient scripts and its long tradition, which has always been a good argument in earlier times. Notably, the amounts to be used are not specified. This must be considered as problematic due to the toxicity of its seeds which may well lead to death (see below).

In total, the Papyrus Ebers describes the following ingredients and indications:

Ingredient(s)	Intended treatment
Root + water	Head
Fruit (chewed up) + beer	Intestinal emptying
Fruit (ground) + oil	Hair growth
Oil from the fruit	Exanthema (treatment over 10 days)
Fruit (rushed) + honey	Pain

Thus, the Papyrus identifies several parts of the plant as useful. Interestingly, albeit nowadays being primarily used, the oil is mentioned only once in the Papyrus Ebers (for external use in exanthema).

Eb 123 (27, 11 - 27, 14): Another, to eliminate pain-matter: Oil prepared from the ricinus plant (dgm). Who suffers from the wb5-w-skin-dis~ase and is burdened with iU.t (and) Wb5'W from which he is ill, is anointed therewith. The rjwm'w stop as in one whom nothing had befallen. He will also be treated (bnm), however, with ointments (wrb) corresponding to the 10 days during which one anoints (wrb) very early, so that they are eliminated. Really effective, a million times.

The final sentence that it has been proven millions of times provides the additional criterion of quantity. Prescriptions based on plants other than ricinus exist as well, but they often rely on unidentified ingredients. In this regard, ricinus oil is the most straightforward prescription considering ingredients and availability. Also, it seems to be particularly effective in exanthema. Another example, this time related to the head:

Eb 437 (64, 14 - 64, 15): Beginning of the remedies to eliminate the bnsj-t-disease on the head: Fruit of ricinus (dgm) 1; (ox) fat 1; behen-oil 1; made into a mass; bandaged (wrb) with it every day.

The fact that many more prescriptions based on other ingredients derived from other plants, animals, or minerals exist suggests the high prevalence of this disease. A final example explores the use of ricinus as an universal analgesic:

Eb 601 (76, 16 - 76, 18): Another, to eliminate the mr-t-disease in any body-part of man: Ricinus (dgm) fruit; pounded (hb!f); added to honey; dressed therewith.

Fig. 1 The text passage (Eb 251) (column 47, line 15 – column 48, line 3) is often named as “Book Ricinus” and summarizes the curative applications of the ricinus plant (see text for details). The red ink is used to indicate headings and important statements as well as to highlight information on quantities (image courtesy of the University Leipzig Library, Papyrus- and Ostraka collection)

The anti-inflammatory effect of honey is well known and broadly used (Manjunatha and Chua 2014). It can only be speculated if this effect is enhanced by the crushed ricinus seeds.

Notably, only a very few and clearly identifiable excipients are employed in the context of ricinus: water, beer, honey, fat, oil, and behen oil/olive oil. This has proven to be very helpful since 70–80% of the ingredients and excipients mentioned in the Papyrus Ebers cannot be identified. Often, the ancient Egyptian art of healing uses rather a mix of *similia* and *contraria* according to the motto “the more, the better,” since remedies are often composed of dubious ingredients according to the principles “*similia similibus curantur*” and “*contraria contrariis curantur*.” It is also interesting to note that religious or magic elements, sometimes found in the Papyrus Ebers, are completely missing in the ricinus prescriptions.

Since the Papyrus Ebers covers remedies, it does not focus on deliberately induced poisoning-despite the widespread fear of poison in ancient times. Thus, only very few prescriptions exist in the Papyrus that are explicitly intended for inducing damage. These are not based on parts of the ricinus plant and are thus beyond the scope of this paper which will rather focus on a description of the plant and its ingredients according to our knowledge nowadays.

The plant *Ricinus communis* and its most important ingredients

Ricinus communis (*Ricinus communis* L.) is a member of the spurge family Euphorbiaceae. The plant is the only species and the genus is monotypic (Buch 2018 and references therein). While originating from wet tropics to subtropical dry regions, it grows well in many areas and on different continents around the world (examples are given in Suppl. Fig. 1). In warm regions, it can reach up to 8–10 m in height; it lives for many years and is perennial. The flowers in the

upper part of the tip of the spikes are female and bear the fruit (Fig. 2a, b, d). The flowers in the lower part are male and wither after yielding their pollen (Scarpa and Guerci 1982) (Fig. 2a, c, d). At the end of the vegetation period, the green to red-brown seed vessels are developed (Fig. 2e). The fruit is formed by three boxes with separate ripe, with each containing one marmorate seed (Fig. 2f, g). The seeds are oval, bean-like, and light brown, mottled with dark-brown spots and different in length (0.5–2 cm; Bradberry et al. 2003), and have a warty appendage called the “caruncle” (Fig. 2g). The leaves, roots, and seeds are utilized in herbal medicine and beyond. After pressing and extraction of the seeds, the toxin-free castor oil is used in a number of industrial processes and products as well as in medical and cosmetic preparations. Castor bean meal, press cake, and other residues of the castor oil production also serve as a protein source for feed or fertilizer.

From *Ricinus communis*, various components have been isolated and identified as toxic, for example, the highly toxic ricin (ricin D, RCA60, RCA-II), the less toxic, but highly homologous *Ricinus communis* agglutinin (RCA120, RCA-I), and the alkaloids ricinine and nudiflorin as well as allergenic compounds (Fig. 3). A number of varieties (cultivars) of *Ricinus communis* exist. Furthermore, the properties of the constituents in the plant and seeds are influenced by the geographical region, season, period of plant growth, the time point of harvest of the beans, or the moisture content (for review, see Hänsel et al. 1994; Audi et al. 2005; Lopez Nunez et al. 2017). Therefore, a somewhat endogenous variability in the “different cultivars” has been described, also with regard to the protein content (e.g., Despeyroux et al. 2000; Sehgal et al. 2010).

Ricin exists in different isoforms, depending on the seed’s type (size, shape) and plant variety from which it is purified (Despeyroux et al. 2000; Griffiths et al. 2007; Sehgal et al. 2010). Ricin A, B, C, D, and E have been described in the literature (for details, see, e.g. Ishiguro et al. 1971a, b; Lin and Liu 1986; Araki and Funatsu 1987). The name ricin refers to

Fig. 2 Examples of parts of the blossom, seed capsulas, and seeds of the plant *Ricinus communis*. **a, d** Examples of the inflorescences. **a, b** The flowers in the upper part of the inflorescences are female, characterized by their red fertile stigma, and finally bearing the fruit. **a, c** The flowers in the lower part (yellow) are male. **d** A special variety of the plant with red

leaves and fruits (ornamental plant, e.g., in parks and gardens). **e** Development of the green (to red-brown) seed capsulas at the end of the vegetation period. **f** The fruit comprises three boxes with separate ripe, each containing one marmorate seed (**g**)

ricin D. Ricin D was purified by gel filtration and cellulose chromatography (Ishiguro et al. 1971a, b). The co-existence of the two highly similar proteins, the cytotoxin ricin D (RCA60) and the hemagglutinin (RCA120), emerged when using improved separation and isolation methods, and by molecular identification of two different genes (Olsnes et al. 1974; Roberts et al. 1985; Lin and Liu 1986).

Fig. 3 Schematic overview of major components of the plant *Ricinus communis*

A number of groups have studied the different ricin isoforms. Sehgal et al. (2010) subfractionated ricin by chromatography into ricin I, II, and III. Molecular weights between 60 and 65 kDa were reported, associated with biological, immunological, and structural differences (Sehgal et al. 2010). Cytotoxicity studies revealed that ricin III is 4–8 times more toxic than the other two forms (Sehgal et al. 2010 and references therein). Leshin and co-workers found that the *Ricinus communis* genome contains genes encoding seven different but closely related proteins, putative members of the preproridin family of ribosome-inactivating proteins, including ricin toxin and other ricin-like proteins (Leshin et al. 2010).

In 1977, the group of Funatsu isolated a toxic protein distinct from ricin D, referred to as ricin E. Toxicity in mice and cytoagglutinating activity in Sarcoma 180 ascites tumor cells were found equal to those of ricin D, and this cytoagglutination was inhibited by galactose (Mise et al. 1977). In a later paper, ricin E was identified as a fusion product derived from the recombination of the ricin D and *Ricinus communis* agglutinin genes (Araki and Funatsu 1987). The study of Despeyroux and co-workers revealed a large extent of heterogeneity between different cultivars (the D form which is found in large grain seeds, whereas the small

grain seeds seem to contain both D and E forms of ricin toxins) and demonstrated that ricin toxins consist of a series of glycosylated proteins most likely originating from a multigene family (Despeyroux et al. 2000).

Additionally, diverse phytochemicals such as alkaloids, flavonoids, terpenes, saponins, phenolic compounds like kaempferol, gallic acid, rutin, lupeol, ricinoleic acid, pinene, thujone as well as gentisic acid have been described as bioactive compounds in *Ricinus communis* (see Abdul et al. 2018 and references therein).

(i) *The leaves* are rich, e.g., in potassium nitrate (Scarpa and Guerici 1982) and ricinine (Waller and Skursky 1972). The separation and determination of the disaccharide glycoside rutin, gentisic acid, quercetin, and gallic acid have been described (Chen et al. 2008). Additionally, the leaves contain a dye that makes a deep blue color (Scarpa and Guerici 1982).

(ii) *The castor beans* are cultivated for their seeds, yielding the viscous, pale yellow, nonvolatile and nondrying castor oil (Patel et al. 2016). Castor oil becomes even more doughy upon contact with air. A very unpleasant taste has often been described as scratching the throat.

The castor oil seeds contain about 45–55% oil and up to 25% protein as well as carbohydrates (e.g., fructose, glucose, saccharose), squalen, succinic acid, bitter compounds, lecithin, and others (Hänsel et al. 1994; Frohne and Pfänder 2004). Other references also report castor oil to contain up to 90% ricinoleic, 4% linoleic, 3% oleic, 1% stearic, and less than 1% linolenic fatty acids (Patel et al. 2016 and references therein).

(iii) *The oily cake of pressed seeds*, generated in large quantities from the process of castor oil production, contains ~3% ricin (Weber 2014) and ricinin as well as allergenic compounds (e.g., castor bean allergenic fraction CB-1A). After oil extraction and detoxification, the defatted press cake is used as low-value feed and as organic fertilizer, e.g., because of its high nitrogen content or the fast rate of mineralization (see Worbs et al. 2011; Severino et al. 2012 and references therein).

The evaluation of extracts of *Ricinus communis* also identified insecticidal properties, e.g., against aphids (Olaifa et al. 1991), tobacco mosaic virus infection (Taylor et al. 1994), *Anopheles arabiensis* or *Culex quinquefasciatus* (Elimam et al. 2009; Wachira et al. 2014), and a number of other activities (see, e.g., Upasani et al. 2003; Zahir et al. 2010; Abdul et al. 2018). Furthermore, the anti-nematode effect of castor meal was found (Severino et al. 2012 and references therein).

Finally, *Ricinus communis* shows an interesting affinity towards heavy metals. For example, it has been described as a hyperaccumulator of lead and an effective accumulator of nickel and, to a lesser extent, of cadmium (for references, see Severino et al. 2012). The growth was inhibited when, e.g., cadmium, lead, copper, nickel, and zinc were added to the soils.

Ricinoleic acid

Cold-pressed *oleum ricini* can contain up to 90% of ricinoleic acid, as triglyceride of the hydroxylated unsaturated fatty acid (IUPAC, (9Z,12R)-12-hydroxyoctadec-9-enoic acid). Castor oil has been used as a purgative and is described as an effective laxative. After oral intake of the castor oil, the triglyceride is hydrolyzed by intestinal lipases into glycerol and ricinoleic acid. It exerts a laxative effect by irritating the intestinal mucosa and induces labor in pregnant females (Teuscher and Lindequist 2010; Tunaru et al. 2012). In addition to effects on intestinal ion transport and water flux, ricinoleic acid can directly influence intestinal motility. Tunaru and co-workers could show that ricinoleic acid specifically activates the prostanoid receptor (EP3) for prostaglandin E2, suggesting a possible role of the EP3 receptor as a target in the induction of laxative effects (Tunaru et al. 2012 and references therein; see Castor oil for more details).

Furthermore, in several models of acute and subchronic inflammation, ricinoleic acid has been shown to exert analgesic and anti-inflammatory effects after topical application (Vieira et al. 2000). Interestingly, pharmacological analyses demonstrated similarities between the effects of ricinoleic acid and those of capsaicin, suggesting a potential interaction of the ricinoleic acid on sensory neuropeptide-mediated neurogenic inflammation. Anti-inflammatory and analgesic properties of some ricinoleic acid derivatives are also highlighted (Pabis and Kula 2016). Finally, ricinoleic acid also acts as a specific algicide for the control of cyanobacteria (formerly called blue-green algae) (US 4398937, 1983).

Ricin (ricin D, RCA-II, RCA60)

Ricin D is stored in vacuoles within the endosperm cells of mature ricinus seeds and is prone to rapid degradation by hydrolysis during the early stages of post-germination growth (see Hänsel et al. 1994; Hartley and Lord 2004; Greenwood et al. 2005; Lord and Spooner 2011; Tyagi et al. 2015; Lopez Nunez et al. 2017 and references therein). While the endogenous function of ricin within the plant remains elusive, it is speculated that its profound cytotoxicity might serve as a defense against all sorts of herbivorous or other plant-damaging organisms (Worbs et al. 2011 and references therein).

The seminal work of R. Kobert and co-workers (University of Dorpat; now University of Tartu, Estonia) on plant toxalbumins has elucidated the toxic principle of *Ricinus communis*. His assistant H. Stillmark described a toxin that caused agglutination of erythrocytes and precipitation of serum proteins and termed it ricin (Stillmark 1888; Kobert 1906).

Nowadays, ricin is classified as a type 2 ribosome-inactivating protein (RIP2) and is characterized as a disulfide-linked heterodimer (AB toxin) of ~62 kDa, comprising a covalently linked A (~32 kDa) and B subunit

(~34 kDa) (Funatsu and Funatsu 1977; Lord et al. 1994). The catalytically active A subunit (A chain; RTA) irreversibly inactivates ribosomes, leading to irreversible inhibition of protein synthesis and eventually resulting in cell death.

The comparison of the protein sequence of RTA with type 1 RIPs revealed generally low identities (Suppl. Fig. 2; Fabbrini et al. 2017). Higher protein sequence identities and homologies are found when comparing ricin and mistletoe lectin (ML-I) from *Viscum album* (Suppl. Fig. 3).

Degenerative alterations were observed following ricin application to feline dental pulps (Henry et al. 1987). The injection of ricin into the rat facial nerve resulted in “suicide transport” and rapid degeneration of facial motor neurons (Streit and Kreutzberg 1988). Ricin is transported retrogradely by the axons to the cell bodies, causing selective destruction of the neurons in the ipsilateral nodose ganglion as well as those in the dorsal motor nucleus (see Ling et al. 1991 and references therein). Sequential degenerative changes of the sensory neurons as well as the accompanying response of the satellite cells were described (Ling et al. 1991). Törnquist and co-worker investigated the response of glial cells and the activation of complement following motor neuron degeneration upon ricin injection into the hypoglossal nerve (Törnquist et al. 1997). Ricin, when injected into skeletal muscles or peripheral nerves, mimics the syndrome of human motor neuron disease since it affects only motor neurons and sensory neurons (de la Cruz et al. 1994). Ricin is also effective in the inhibition of the protein synthesis in vitro in cell cultures enriched for microglial cells, astrocytes, or cerebellar granule neurons. The glial cells, in particular microglia, are very sensitive to RIP toxicity (ricin, volkensin) and are hypothesized to be a primary target of these toxins when injected in vivo (Sparapani et al. 1997).

***Ricinus communis* agglutinin (RCA120)**

Beyond ricin, the seeds also contain *Ricinus communis* agglutinin (RCA120; ~120 kDa, comprising two A (~32 kDa) and B subunits (~36 kDa)), respectively. The homology between RCA120 and ricin (RCA60) is over 93% between the A chains and 84% between the B chains (Roberts et al. 1985). Amino-terminal sequencing of ricin and RCA120 has shown that the A subunits are identical up to residue 7 and the B subunits are homologous at least in the first 19 residues (see Butterworth and Lord (1983) and references therein). Albeit being highly homologous to ricin, it is categorized as a protein with considerable lower toxicity. Unlike ricin, RCA120 is not directly cytotoxic but shows affinity for red blood cells, leading to agglutination and subsequent hemolysis (see Audi et al. 2005; Severino et al. 2012 and references therein). *Ricinus communis* agglutinin is not significantly absorbed from the gut, thus causing clinically significant hemolysis only after intravenous administration (Balint 1974; Audi et al. 2005).

Ricinine

Ricinine (IUPAC: 4-methoxy-1-methyl-2-oxo-1,2-dihydropyridin-3-carbonitrile) is an alkaloidal toxin (molecular weight = 164.2 g/mol), belonging to the group of piperidine alkaloids. It is found in small amounts in all parts of the plant, including the leaves and the pericarp (e.g., the seeds contain approximately 0.2% of the alkaloid) and translocates in the plant depending on the age (Waller and Skursky 1972; see Audi et al. 2005; Worbs et al. 2011 and references therein). LD₅₀ values for ricinine (mice) were determined at 3 g/kg upon oral incorporation and 340 mg/kg upon intraperitoneal application (Ferraz et al. 1999).

Ricinine is stable in human urine when heated up to 90 °C for 1 h as well as upon storage at 25 °C, 5 °C, or –20 °C for 3 weeks (Johnson et al. 2005). Furthermore, unlike ricin, ricinine cannot be inactivated by conventional heat treatment due to its high temperature resistance (melting point ~200 °C) (Worbs et al. 2011). Since ricinine can be coextracted with ricin, it can be used as a surrogate marker for tracking intoxications caused by crude plant extracts (Darby et al. 2001; Lopez Nunez et al. 2017; Robert Koch Institut 2017).

In animal models, *Ricinus communis* extracts exert typical CNS-stimulating and neuroleptic effects, whereas ricinine has been described to elicit hyperreactivity, clonic seizures accompanied by electroencephalographic alterations, and subsequent death (Ferraz et al. 1999, 2002). Data of Ferrenz and co-workers suggest that an increased release of glutamate in the cerebral cortex can be implicated in the genesis of ricinine-induced seizures and that it triggers many anticonvulsive mechanisms, like the release of Tau, dopamine, serotonin, and noradrenaline (Ferraz et al. 2002).

The powdered leaves of the plant have been used for repelling aphids, mosquitoes, white flies, and rust mites, probably due to the presence of the alkaloid ricinine (Olaifa et al. 1991; Rana et al. 2012). In fact, the alkaloid was described to act as a repellent or a toxin against insects, e.g., the leaf-cutting ant (*Atta sexdens rubropilosa*) or a noctuid moth (*Spodoptera frugiperda*) and has therefore attracted interest with regard to crop protection (Bigi et al. 2004; Cazal et al. 2009; Lopez et al. 2010).

Allergenic compounds

Pollen and seed components of *Ricinus communis* can exhibit an allergenic potential, associated with urticaria as well as conjunctivitis or rhinitis (Bradberry et al. 2003; Teuscher and Lindequist 2010; Deus-de-Oliveira et al. 2011; Severino et al. 2012). Beyond laboratory personnel, castor bean allergies have been found in various industrial sectors, namely in workers employed in fertilizer retail, in the upholstery industry, or in oil-processing mills (Deus-de-Oliveira et al. 2011). Regarding the latter, endemic asthma caused by castor bean dust has been observed in the proximity of a castor oil mill,

and workers occupationally exposed to castor bean dust exhibited the allergic syndrome (see Bradberry et al. 2003; Severino et al. 2012; Patel et al. 2016 and references therein).

Both type I and IV allergenic responses have been described following dermal exposure to castor bean dust, as well as the involvement of eosinophils and IgE receptors (Bradberry et al. 2003 and references therein). Spies and Coulson isolated a protein fraction and called it CB-1A (Spies and Coulson 1964). CB-1A belongs to a family of low molecular weight storage proteins named 2S albumins that is present in seeds of a wide range of dicotyledonous plants (for references, see Severino et al. 2012). Specific IgE antibodies against the 2S storage albumins have been detected in most (96%) castor-sensitive patients (Thorpe et al. 1988; Deus-de-Oliveira et al. 2011). Other isoforms of allergenic 2S albumins were determined and named Ric c 1 and Ric c 3, and critical amino acids in the IgE-binding epitopes in these isoforms have been identified (Deus-de-Oliveira et al. 2011; Severino et al. 2012 and references therein). It has been suggested that these castor allergens Ric c 1 and Ric c 3 are a new class of amylase inhibitors and may play a role in plant defense, based on their inhibition of insect α -amylase (Nascimento et al. 2011).

Ricinus lipase

The castor bean exhibits powerful lipase activity, which is responsible for the hydrolysis of triglycerides (Srivastava et al. 2016). Lipase activity is localized in the spherosomes derived from endosperm tissue of the seeds and, in contrast to other oilseeds, the castor bean lipase is activated in dormant seeds and in germinated seeds (Ory et al. 1968; Srivastava et al. 2016). More specifically, two lipases have been described in extracts from castor bean endosperm, one with optimal activity at pH 5.0 (acid lipase; associated with the membranes of spherosomes) and the second with an alkaline pH optimum (alkaline lipase; associated with the membrane of glyoxysomes and the endoplasmic reticulum) (Muto and Beevers 1974; Eastmond 2004; Srivastava et al. 2016). Furthermore, Morlon-Guyot and co-worker have shown that ricin possesses a functional lipase active site at the interface between the two subunits. The structural examination of type II (synonymous to “type 2” as found in other references) RIPs indicated that this lipase site is present in toxic (ricin, abrin) members of this family, but that it is altered in lesser toxic or nontoxic (mistletoe lectin I, ebulin 1) members (Morlon-Guyot et al. 2003). This finding further indicates that lipolytic activity likely plays an important role in RIP cytotoxicity.

Nowadays, ricinus lipase derived from press cake is employed in industrial fat processing. Notably, lipases represent one of the most abundantly described groups of enzymes in the context of biofuel production. More specifically, it is

used for the processing of glycerides and fatty acids for biodiesel (fatty acid alkyl esters) production (Ribeiro et al. 2011).

Mechanisms of ricin action

As described above, ricin is a heterodimer (AB toxin) consisting of a sugar-binding B chain (~34 kDa; 262 aa) linked via a single disulfide bond to the catalytically active A chain (~32 kDa; 267 aa). This results in a holotoxin of about 65 kDa (Lappi et al. 1978; Lord and Spooner 2011; Worbs et al. 2011 and references therein). The mechanism of action and toxic principle of ricin have been studied among others by Endo and co-workers, characterizing ricin (and other plant toxalbumins) as RNA N-glycosidases (Endo and Tsurugi 1987; Endo et al. 1987). Notably, ricin-related toxins like abrin and modeccin exert similar activities on 28S rRNA, suggesting a more general mechanistic pathway for ribosome inactivation by lectin toxins (Endo et al. 1987).

Ricin preferentially binds through the catalytically inactive B chain, a lectin with galactose- and N-acetylgalactosamine specificity, to carbohydrate residues on the cell surface, thus facilitating the internalization of ricin into the cytosol (for a schematic illustration, see Fig. 4). In fact, several oligosaccharide residues including N-acetylglucosamine and galactose residues on glycolipids and glycoproteins, which are broadly present on mammalian cells, have been identified as receptors for the lectin subunit (Worbs et al. 2011). Additionally, a mannose receptor (MR; CD206)-mediated uptake of the ricin toxin has been described (Simmons et al. 1986; Magnusson et al. 1993; Gage et al. 2011).

Ricin is internalized through clathrin-dependent and clathrin-independent endocytosis. It undergoes retrograde transport from endosomes to the Golgi apparatus towards the endoplasmic reticulum (ER). In the ER, a disulfide isomerase mediates the cleavage of the disulfide bond between the A and B chain, allowing the cytotoxic A chain to translocate across the membrane of the ER into the cytosol (Lappi et al. 1978; Lord and Spooner 2011; Sandvig et al. 2013; Tyagi et al. 2015 and references therein). Here, through removal of a single adenine residue from the so-called sarcin loop of the eukaryotic 28S rRNA (position 4324; leading to depurination), the A chain irreversibly inactivates eukaryotic ribosomes and thus inhibits protein synthesis (Lord et al. 1994; Hartley and Lord 2004; Tyagi et al. 2015). On the mechanistic side, this involves the hydrolytic cleavage of the N-glycosidic bond of A4324 of the 28S rRNA (Endo et al. 1987; Endo and Tsurugi 1987). This adenine, however, is the binding site for elongation factors 1 and 2, and the depurination profoundly impairs the ribosomal elongation cycle, rendering these ribosomes incapable of binding translation factors and thus abolishing protein synthesis. This results in cell death, thus accounting for the extreme cytotoxicity of

Fig. 4 Schematic illustration of the mechanism of ricin internalization and action. Ricin binds to cell surface receptors by its ricin B chain, prior to internalization via endocytosis. Subsequent processes are lysosomal degradation or exocytosis, or transport to the Golgi complex and then, by a retrograde transport mechanism, to the endoplasmic reticulum (ER). In the lumen of the ER occurs the cleavage of ricin A and B chains. In the cytosol, the ricin A chain inactivates ribosomes by depurination of 28S rRNA in the ribosomal subunit (adenine at position 4324 of the α -sarcin/ricin loop). Artwork by courtesy of Dr. Jens Grosche (Effigis AG)

ricin (for review, see Lord and Spooner 2011). Functional domains like the α -helix formed by residues 99–106 have been identified as instrumental in the control of ricin's depurination activity (Dai et al. 2011). Finally, Taubenschmidt et al. discussed a potential interplay between fucosylation, galactosylation, and sialylation with regard to ricin toxicity, since they identified for the first time an extremely high specificity of ricin for defined glycosidic structures that determine the cellular fate upon ricin exposure (Taubenschmidt et al. 2017).

Apart from protein synthesis inhibition, other cytotoxic mechanisms have been put forward (examples of important cellular studies on ricin are listed in Table 1 and examples of the important toxic effects of ricin are shown in Suppl. Fig. 4). These include electrolyte imbalance, damage to cell membranes or alterations in their structure and function, the release of cytokines as mediators of inflammation, hepatic oxidative stress, and the direct induction of apoptosis (Audi et al. 2005; Worbs et al. 2011; Tyagi et al. 2015; Lopez Nunez et al. 2017). Consequently, in animal studies elevated plasma ALT and AST levels were found upon administration of ricin A chain, indicating hepatotoxicity and increasing inflammatory responses (Buonocore et al. 2011). Finally, it has also been shown that seeds, stem, leaves and root methanolic extracts from *Ricinus communis* induced mild to moderate cytotoxicity against human or bovine red blood cells and also exerted mild mutagenicity (Abbas et al. 2018).

Strikingly, only a single ricin A-chain molecule is sufficient for inactivating >1500 ribosomes per minute, and thus to induce cell death (Audi et al. 2005). A number of studies aimed at influencing these mechanisms for the specific manipulation

of the ricin activity (see below). Among others, this has led to the identification of a “druggable” sugar code that is evolutionary conserved and can be manipulated for controlling toxicity of ricin (Taubenschmidt et al. 2017).

Ricin toxicity

Ricin is one of the most toxic plant toxins. The ricin content can be up to 1–5% (w/w) of the beans of the castor oil plant (Balint 1974; Bradberry et al. 2003). Precision and reproducibility of its toxicity are influenced by a considerable number of factors:

(i) As described above, ricin dosages and toxic effects estimated from the number of beans ingested may be inaccurate due to the plant varieties and variations in region-dependent growth conditions as well as the quality of the beans.

(ii) The features and severity of ricin toxicity/poisoning vary markedly with the dosage and route of exposure/uptake (oral, inhalative, per injection, dermal) and furthermore, on the individual's side, from the degree of mastication, age, and comorbidities (for review, see Bradberry et al. 2003; Audi et al. 2005).

(iii) When characterizing ricin toxicity (kinetics, esp. distribution and excretion) in the lab, results also depend on the experimental system used, e.g., cell culture conditions and assays in vitro, animal species, strains, age, sex, and feeding conditions used in vivo as well as the injection/intake route and observation time (e.g., Roy et al. 2012).

Thus, extrapolating results from animal experiments towards humans remains difficult. Still, the following numbers are often cited (see also Table 2): (i) The lethal dose by inhalation (breathing in solid or liquid particles) and injection (into

Table 1 Examples of important cellular studies on ricin

Cell system	Cellular effects	Molecular effects	Comments/consequences	References
Human endothelial cells	Changes in morphology; ↑permeability of umbilical vein endothelial cells	- Endothelial cells bind 125I-rRTA in a dose-dependent manner	- In vitro model for vascular leak syndrome	Soler-Rodríguez et al. (1993)
Rabbit coronary arteries	Contraction and relaxation of coronary arteries; alterations in coronary arteries	- Responses to 5-HT, histamine, NE, ACh - Cumulative contractions of rabbit coronary artery rings to 5-HT and histamine - Cumulative relaxations to ACh and NE were measured in rings - ↑EC ₅₀ of contractions of coronary artery rings to 5-HT and histamine - ↓EC ₅₀ of relaxation of coronary artery to NE	- ↑Cardiac output and blood flow - Causes hemorrhage and necrosis in the heart, following a lethal dose - Hypothesis: (i, early stage of ricin intoxication): the sum of the effects is to reduce the vascular tone of the coronary artery and thus reduce blood pressure, (ii, late stage of ricin intoxication): vasospasm of coronary artery may cause micro- and macrocirculatory collapse	Zhang et al. (1994)
Isolated rabbit heart	Cardiac function	- Ricin reduced both systolic (left ventricular developed pressure and +dp/dt) and diastolic (compliance) left ventricular functions, perhaps due to increased vascular permeability	- Binding studies: ricin neither reduced beta-adrenergic receptor numbers nor altered the dissociation constant - Ricin effects perhaps due to increased vascular permeability, without altering responses to the alpha- and beta-adrenoceptor agonists	Ma et al. (1996)
Microglia, astrocyte, and neuron cultures	Toxicity; inhibition of protein synthesis	- Toxicity (lactate dehydrogenase release) - Inhibition of protein synthesis (similar potency in cerebellar granule neurons and astrocytes)	- Glial cells (in particular microglia) are very sensitive to RIP toxicity	Sparapani et al. (1997)
Human lens epithelial cell	Cytotoxicity; protein synthesis and proliferation	- Inhibition of protein synthesis - Inhibition of proliferation on the inner surface of the lens capsule	- Immunotoxin 4197X-RA may help to prevent posterior capsule opacification after primary cataract surgery	Tarsio et al. (1997)
Hepatoma cells (BEL7404)	Mechanism of cytotoxicity; induction of apoptosis	- Bcl-2 could promote the survival of the hepatoma cell against ricin insult - Induced apoptosis was accompanied by ↑Bak, ↓Bcl-x, ↓Bax expression - Caspases and poly (ADP-ribose) polymerase cleavage activity are implicated in cell death	- Calcium-dependent proteases or PKC do not participate in the apoptotic process - Ricin exerted its cytotoxicity through nonspecific inhibition of de novo protein synthesis in general	Hu et al. (2001)
Monocyte-derived dendritic cells (DC)	Lack of dendritic cell maturation; ricin variants retain significant ribotoxicity and catalytic activity	- Ricin neither induces DC maturation nor interferes with LPS-induced DC maturation (in contrast to the bacterial proteins) - No correlation between the absence of DC maturation and ricin dysfunction	- Understanding the effect of ricin may provide insights into the generation of an anti-ricin vaccine and into the use of inactivated ricin A chains as delivery vectors as part of a vaccination protocol	Smith et al. (2004)
RAW 264.7 macrophages	Induction of a severe inflammatory response	- Activation of ERK, JNK, p38 MAPK - Accumulation of mRNA encoding TNFα, IL-1, c-Fos, c-Jun, EGR1 - TNFα protein in culture medium	- Therapeutic management of inflammatory response may affect the outcome of intoxication by ricin	Korcheva et al. (2005)
Primary murine bone marrow derived-(BMDM) and alveolar macrophages (AM)	Cross talk between apoptotic and inflammatory signaling pathways	- Release of pro-inflammatory cytokines - Transcriptional gene activation - AM: activation of stress-activated protein kinases; ↑expression of pro-inflammatory mRNA transcripts; ↑synthesis and secretion of TNFα	- Macrophages (considered as upstream regulators of inflammatory cascades) may play a central role in pathogenesis and development of ricin-induced ARDS because of their ability to secrete pro-inflammatory cytokines	Korcheva et al. (2007)
Murine bone-marrow derived macrophages	IL-1β release	- Apoptotic cell death - Dependent on the NALP3 inflammasome - Suppressed by ROS	- IL-1β-dependent inflammatory signaling	Lindauer et al. (2010)

Table 1 (continued)

Cell system	Cellular effects	Molecular effects	Comments/consequences	References
NIH 3T3 cells	Induction of apoptosis	- Activation of caspase 3 and caspase 8, but not caspase 9 - Different in vitro toxicity of structurally similar type I RIPs	- Cell death mechanism may vary depending on the experimental conditions (necrosis in vivo and apoptosis in vitro)	Sha et al. (2010)
Rat sensory neuronal and Schwann cell cultures	Induction of apoptosis	- Strong apoptotic signal in neurons - Attenuated apoptotic reaction (Schwann cells)	- All internalized RIPs were found concentrated in the cellular cytoplasm	Tong et al. (2012)
Murine bone marrow derived macrophages	Suppression of ribosomal function, triggering innate immune signaling	- Activation of the NLRP3 inflammasome - Synthesis of pro-IL-1 β , release of IL-1 β	- IL-1 β -dependent inflammation through activation of the NLRP3 inflammasome	Vyleta et al. (2012)
Mouse macrophages RAW264.7	Macrophage activation	- Increase in iNOS, TNF α , IL-6 - Transduction mechanisms: PI3K, PKC, JAK2, p38, MAPK, NF- κ B	- Proteins are associated with the downstream cascades of activated JAK-STAT and NF- κ B receptors	Xu et al. (2013)

Abbreviations: *ACh* acetylcholine; *ARDS* acute respiratory distress syndrome; *JNK* c-Jun N-terminal kinase; *iNOS* inducible nitric oxide synthase, (IL) interleukin; *JAK/STAT* Janus kinase/signal transducers and activators of transcription; *LPS* lipopolysaccharide; *MAPK* mitogen-activated protein kinase; *NALP3* (or NLRP3/cytopyrin) inflammasome; *NE* norepinephrine; *NF- κ B* nuclear factor- κ B; *PI3K* phosphatidylinositol 3-kinase; *PKC* protein kinase C; *ROS* reactive oxygen species; *5-HT* serotonin; *TNF* tumor necrosis factor

muscle or vein) has been estimated to be approximately 5–10 μ g/kg body weight in man, i.e., 350–700 μ g for a 70-kg adult (Bradberry et al. 2003). (ii) When analyzing cases of poisoning upon castor bean ingestion, the lethal oral dose for ricin in humans has been estimated at ~1–20 mg ricin/kg body weight (equivalent to approximately 8 beans, Audi et al. 2005). In reports documenting clinical symptoms (mild to lethal), the number of beans ingested range from 0.5–30 (Challoner and McCarron 1990; Audi et al. 2005; Worbs et al. 2011). Concomitantly, while it has been reported that up to 1 mg pure toxin can be isolated from 1 g seeds, representing a lethal dosage, other references rather describe 1–6 or 10–20 seeds to be toxic in children or adults, respectively (see Roth et al. 2008; Thiermann et al. 2013 and references therein).

For comparison, the LD₅₀ values in mice are ~3–24 μ g/kg after injection or inhalation and ~20–30 mg/kg after castor seed ingestion—which is approximately 1000-fold higher (see also Table 3; e.g., Franz and Jaax 1997; He et al. 2010; Audi et al. 2005, Worbs et al. 2011 and references therein). A number of case reports on intoxication have been published, including the prominent umbrella

attack on the Bulgarian dissident Georgi Markov (killed in 1978 with the tip of an umbrella while waiting for a bus in a London street; Crompton and Gall 1980). Very comprehensive summaries of ricin intoxications in humans due to castor seed ingestion are given by Worbs et al. (2011) and Roxas-Duncan and Smith (2012).

Ricin intoxication

Ricin is available, e.g., as crude impure plant extract, purified crystals, powder, and solution. Purified ricin is a white powder that is soluble in water and stable under ambient conditions. Heating for 10 min at 80 °C or for 1 h at 50 °C (pH 7.8) may lead to its inactivation (Burrows and Renner 1999; Audi et al. 2005; Vitetta et al. 2006).

The effects of ricin (clinical symptoms) are directly related to the route of administration: oral, parenteral, inhalative, or dermal. Furthermore, different individual factors of the patients are also responsible for the progress and the symptomatology (Robert Koch Institut 2017).

Table 2 Examples of toxicological data estimated for ricin in humans

Exposure	Species	Toxicity/LD	Reference
Inhalation, injection (i.m. or i.v.)	Human	Lethal dose approximately 5–10 μ g/kg (~350–700 μ g in a 70-kg adult)	Bradberry et al. (2003)
Oral	Human	Lethal oral dose approximately 1–20 mg/kg (~8 beans)	Audi et al. (2005)
Inhalation	Human	Lethal dose approximately 3 μ g/kg	CDC* https://www.awl.ch/heilpflanzen/ricinus_communis/CDC_ricin_protocol.pdf
Injection (Georgi Markov)	Human	Fatal dose suggested to be in the order of 1–10 μ g/kg	Crompton and Gall (1980)

*Centers for Disease Control and Prevention

Table 3 Examples of toxicological data for ricin and ricinus agglutinin, as estimated from animal studies

Exposure	Species	Toxicity/LD	Reference
Oral	Mouse	LD ₅₀ is 30 mg/kg body weight	Audi et al. (2005)
Oral	Rat, mouse	Lethal 30 mg/kg body weight	Ishiguro et al. (1983)
Injection (i.v.)	Mouse	LD ₅₀ is approximately 55–65 ng	Fodstad et al. (1976)
Injection (i.p.)	Mouse	(i) LD ₅₀ values were 4, 28, and 14 µg/kg body weight for ricins A, B, and C (ii) LD ₅₀ (ricinus agglutinin) is 112 µg/kg body weight	Lin and Liu (1986)
Injection (i.v.)	Mouse	LD ₅₀ (ricin) is 36.23 ± 6.17 µg/kg LD ₅₀ (ricinus agglutinin) is 1.36 ± 0.56 mg/kg	Zhan and Zhou (2003)
Injection (i.p.)	Mouse	LD ₅₀ is 0.012 mg/kg	Lin et al. (1970)
Injection (i.p.)	Mouse	LD ₅₀ is 0.2 µg/kg	Olsnes and Pihl (1973a)
Inhalation	Rat	(i) LD ₅₀ is 3.7 µg/kg (RC var. Hale Queen*) (ii) LD ₅₀ is 9.8 µg/kg (RC var. zanzibariensis*)	Griffiths et al. (1995)
Inhalation	Mouse	LD ₅₀ (particle sizes less than 5 µm) is ~3–5 µg/kg	Roy et al. (2003)
(i) Inhalation (ii) Injection (i.v.) (iii) Injection (i.p.) (iv) Injection (s.c.) (v) Intragastric administration	Mouse	(i) LD ₅₀ is given as 3–5 µg/kg body weight, with a mean time to death of 60 h (ii) LD ₅₀ is 5 µg/kg body weight and death within 90 h after intravenous injection (iii) LD ₅₀ is 22 µg/kg body weight and death within 100 h after intraperitoneal injection (iv) LD ₅₀ is 24 µg/kg body weight and death within 100 h after subcutaneous injection (v) LD ₅₀ is 20 mg/kg body weight and death within 85 h after intragastric administration	Franz and Jaax (1997)
(i) Oral (ii) Injection (i.p.)	Mouse	(i) LD ₅₀ is 28.29 mg/kg (ii) LD ₅₀ is 1.01 µg/kg	Kumar et al. (2004)
(i) Injection (i.p.) (ii) Injection (i.v.)	Mouse	(i) LD ₅₀ is 110 ng (ii) LD ₅₀ is 88 ng	Foxwell et al. (1985)
(i) Oral/intragastric (ii) Injection (i.p.) (iii) Injection (i.v.)	Mouse	(i) LD ₅₀ is 29 mg/kg (ii) LD ₅₀ is 0.023 mg/kg (iii) LD ₅₀ is 0.008 mg/kg	He et al. (2010)
(i) Oral (ii) Injection (i.p.)	Mouse	(i) LD ₅₀ is 5 to 20 mg/kg body weight (ii) LD ₅₀ is 2 to 20 µg/kg body weight	Garber (2008)
(i) Oral (ii) Pulmonary	Rat	(i) LD ₅₀ is 8 mg/kg body weight (ii) LD ₃₀ is 0.8 µg/0.1 ml	Cook et al. (2006)

**Ricinus communis* variety

The therapy of ricin intoxication is symptomatically. The standard procedure is a rapid primary elimination of the toxin (asservation and control of the stomach contents). The prompt treatment with supportive care is necessary to limit morbidity and mortality. It is important to note that in pre-clinical studies the passive immunization with anti-ricin neutralizing antibodies has been identified so far as the only post-exposure measure effective against pulmonary ricinosis at clinically relevant time points following intoxication (Gal et al. 2017). The efficacy of this anti-toxin treatment depends on the antibody affinity and the time of treatment initiation within a limited therapeutic time window.

An overview of the four ways of intoxication and some characteristics is given in Table 4 and briefly discussed in the following.

Oral intoxication

(i) Routes of intake: Food and water contamination; castor beans found in jewelry (necklaces and bracelets); unintended uptake by children due to the bean's resemblance to nuts, i.e., children were observed eating castor beans because of their attractive, hazelnut-like appearance and the typical taste of the seeds.

(ii) Further remarks: Ricin toxin is not released from the bean until it has been chewed and digested, thus allowing ricin from the formerly intact seed capsule with little toxicity into the gastrointestinal tract. Intoxication depends on the degree of the crushing and from the stomach contents (Roy et al. 2012; Robert Koch Institut 2017). After oral ingestion, ricin is stable towards gastrointestinal proteases and is absorbed from the gastrointestinal tract (Bradberry et al. 2003; Balint 1974). Release of toxin from beans is generally in the ileum or colon (Roy et al. 2012).

Table 4 Overview of intoxication routes and symptoms in humans

Oral intoxication: clinical symptoms	
Latency period	<p>Short (first symptoms (fever, thirst) after 6 h (to 4 days))</p> <ul style="list-style-type: none"> • Gastrointestinal symptoms: nausea, vomiting, diarrhea, abdominal pain, colics necrosis in the walls of the gastrointestinal tract • Loss of fluids and electrolytes; exsiccosis • Muscle pain (rhabdomyolysis) • Nephritis, anuria; liver and kidney damage • Thromboses • Necrosis in the spleen and in the lymphatic system • Spasms in the extremities, cerebral seizures, tremor, tonic-clonic seizures • Vision problems, mydriasis • Circulatory failure <p>Severe intoxication:</p> <ul style="list-style-type: none"> • Mydriasis, seizures, hepatic and renal failure • Circulatory failure • Hemorrhagic gastroenteritis with necrosis
Death	<ul style="list-style-type: none"> • After 3–4 days (without medical therapy) • Death through apnea and cardiac failure
Parenteral intoxication: clinical symptoms	
Latency period	<ul style="list-style-type: none"> • Immediate strong pain at the injection site • Additional symptoms within 24 h (e.g., fever, nausea, vomiting) • Circulatory failure (within a few hours) • Severe general feeling of illness, possibly in combination with fever, headache, fall in blood pressure, higher cardiac frequency, nausea, vomiting, muscle cramps • From muscle pain to necrotic myositis • Cardiac arrhythmias • Shock • Hepatic and renal failure with secondary metabolic implications • Multiorgan failure (collapse) • Agglutination of the erythrocytes • Changes in laboratory parameters, e.g., leukocytosis, (hemolytic) anemia, increase in hepato-specific transaminases, rhabdomyolysis, (lactate-) acidosis, uremia, electrolyte disorders
Death	<ul style="list-style-type: none"> • After 36–48 h • After injection: death through organ or circulatory failure
Inhalative intoxication: clinical symptoms	
Latency period	<ul style="list-style-type: none"> • Rapid accumulation of fluid in the lungs • Short pulmonal symptoms (cough, dyspnea) within 4–8 h • Pulmonary and circulatory failure within 18–24 h • Clinical symptoms as after chemical burn (some hours after aerosol exposure; irritation of the buccal and nasal mucosa) • Strong respiratory symptoms, perturbation of the pulmonary function, bronchospasm, necrotic pneumonia (with alveolar inflammation), lung edema, acute respiratory distress syndrome (ARDS) with fibrotization • Urticaria and severe inflammation (e.g., leukocytosis) • Conjunctivitis, swelling of the mucosa in the oropharynx and in the upper respiratory tract • Respiratory allergies (immediate-type allergy)
Death	<ul style="list-style-type: none"> • After 36–72 h • Respiratory failure and circulatory failure
Dermal intoxication: clinical symptoms	
Latency period	<ul style="list-style-type: none"> • Unknown • Allergic-toxic reaction, strong sensibilization, urticaria (pruritis, erythema, swelling, vesication) • Pain; skin and mucosal irritation or injury • Eye: local injury, inflammation of the eyes

Parenteral intoxication

(i) Routes of intake: Stab wounds, injection (i.m. or s.c., among others), incisions. After application, the toxin is systemically distributed in the body, followed by a severe disease process and impairment of body functions (Robert Koch Institut 2017).

(ii) Further remarks: Ramsden and co-workers studied the distribution and excretion of ricin in rats after intravenous injection of [125 I]-labeled ricin (equal in toxicity to native ricin) (Ramsden et al. 1989). After 30 min, they identified the liver as the major organ of localisation (46% of injected dose), with the spleen and muscle being next (9.9% and 13%, respectively) while levels in lymph nodes were found very low (1.2% / g). Ricin was quickly cleared from the animal; only 11% of the initial radioactivity was left after 24 h, while 70% was excreted in the urine (Ramsden et al. 1989).

Inhalative intoxication

(i) Routes of intake: Ricin poisoning by inhalation as aerosol (ricin powder/ricin dusts; ricin solutions, grist (fertilizer)), as observed in the proximity of ricin mills. Ricin dusts were found to induce immunological reactions in exposed individuals, e.g., in the periphery of factories producing castor oil.

(ii) Further remarks: The toxic effect of ricin after inhalation (aerosol) depends on various factors including the humidity at the time point of exposure, injuries of the lung, and particle sizes of the aerosol, determining the penetration depth into the lung (Audi et al. 2005; Griffiths 2011; Roy et al. 2012). Particles of low micron size were found to deposit deeper in the respiratory tract, resulting in higher mortality (Griffiths et al. 1995; Audi et al. 2005). In contrast, larger particles typically deposit higher in the airways, and can thus be swept out by the mucociliary system and subsequently swallowed (Roy et al. 2012).

Dermal intoxication

(i) Routes of intake: Skin exposure of ricin meal/grist used as fertilizer or in the area of ricin mills leads to allergic reactions. Intoxication is also possible through the conjunctiva.

(ii) Further remarks: Biochemical studies have demonstrated lipophilic properties of ricin, leading to dermal absorption. However, no systemic distribution was found, indicating that this intoxication route as less relevant. In fact, in the case of intact skin, no examples for systemic intoxication have been described beyond the strong sensibilization potential, while after skin injuries local dermal intoxication can be observed.

From past to present-pharmacological and technical use

Beyond castor oil, a number of its phytochemical ingredients have been studied for their pharmacological and therapeutic effects. These include anti-microbial, anti-fungal, anti-cancer, anti-diabetic, anti-inflammatory, anti-malaria, anti-oxidant, central analgesic, anti-convulsant, anti-nociceptive, anthelmintic, anti-fertility, laxative, uterine contracting, anti-implantation, anti-asthmatic, bone regenerative, molluscicidal, anti-ulcer, anti-histaminic, wound-healing, insecticidal, anti-arthritic, anti-dandruff, and hepatoprotective activities (see Hänsel et al. 1994; Abdul et al. 2018). Furthermore, a novel fusion protein between ricin A chain (RTA) and pokeweed anti-viral proteins (PAPs; *Phytolacca Americana*) has been developed. This “RTA-PAPS1” possesses significantly increased inhibitory effects on protein synthesis and anti-hepatitis B virus (anti-HBV) activity in vitro and merits further development as a potentially potent anti-viral agent for the treatment of chronic HBV infection (Hassan et al. 2018).

Castor oil-technical use

Castor oil has long been used in different ways, e.g., as an inexpensive fuel for oil lamps as already described in the Papyrus Ebers. Nowadays, castor oil production is the most important use of the ricinus plant. The castor seeds contain 40–50% oil. Between 2009 and 2013, the average annual world production of castor oil seed was approximately 1.99×10^6 tons (Mensah et al. 2018) and represents ~0.15% of the total vegetable oil produced worldwide (Severino et al. 2012).

Cold or hot pressing of the beans, followed by solvent extraction, allowed for the production of different kinds of oils and lubricants. Castor oil offers a number of advantages, making it an important feedstock utilized by industry (see Suppl. Fig. 5). These include, for example, the high proportion of the fatty acid ricinoleic acid as well as the presence of a number of functional groups useful for performing a variety of chemical reactions (including halogenation, dehydration, alkoxylation, esterification, sulfation; Ogunniyi 2006; Patel et al. 2016; Mensah et al. 2018). For a more detailed description and critical appraisal of the potential of castor oil in the context of biotechnology and industrial uses, including renewable fuels, the reader is referred to the literature (e.g., Mutlu and Meier 2010; Severino et al. 2012; Mensah et al. 2018).

The processed or refined castor oil is devoid of all toxic proteins and can thus be safely used for pharmacological and pharmaceutical applications (in particular, the toxic ricin is insoluble in oil and any residual ricin is thus eliminated during the refining process). In conventional, alternative, and folk medicine, castor oil is still in use as a stimulant laxative. It is generally recognized and classified as safe by the Food and Drug Administration (FDA). The Joint Food and Agriculture

Organization (FAO)/World Health Organization (WHO) Expert Committee on Food Additives established an acceptable daily castor oil intake (in humans) of up to 0.7 mg/kg body weight (Final report on the safety assessment of *Ricinus communis* (Castor) Seed Oil (2007)). Yet, the clinical use of castor oil as a laxative has been decreasing in conventional medicine, while it is still used widely in alternative and folk medicine (Tunaru et al. 2012; Patel et al. 2016 and references therein). However, due to its very strong effects possibly leading to abdominal cramps and its unpleasant taste, it is nowadays considered as obsolete.

On the mechanistic side, the laxative effects of ricin oil were found to be based on their triacylglycerols of ricinoleic acid contents. While the triglyceride is inactive, it is hydrolyzed by lipases in the small intestine, releasing ricinoleic acid. By triggering the release of prostaglandin E₂, it leads to enhanced intestinal motor activity, inhibition of water resorption, and thus increased water and electrolyte secretion into the gut. Tunaru and co-workers reported on the mechanism by which castor oil exerts its effects on gut and uterus motility, inducing laxation and uterus contraction (see also part Ricinoleic acid). They could show that ricinoleic acid is a selective agonist of prostaglandin E receptors (EP₃ and EP₄), mediating the pharmacological effects by activation of EP₃ receptors on smooth-muscle cells. The authors verified that EP₃ and EP₄ receptors are expressed in MEG-01 cells and that prostaglandin E₂ has effects comparable to ricinoleic acid (Tunaru et al. 2012). These results also suggested EP₃ receptors as potential targets for drugs to induce laxation. For research purposes, castor oil has also been administered orally to induce diarrhea in rats. This has led to the establishment of a bioassay for the measurement of anti-diarrhea activity, representing a fast and efficient method for preliminary screening for potential drug-like candidates from natural products (for references, see Patel et al. 2016).

Finally, castor oil and hydrogenated castor oil are used as ingredients in cosmetics, e.g., in lipsticks or hair care products; as supplement in deodorants (zinc ricinoleate), hand creams, and lotions; or in products for foot care. Refined ricinus oil is employed for the purpose of injections and in eyedrops (Final report on the safety assessment of *Ricinus communis* (Castor) Seed Oil (2007); Arnold 2017).

In modern medicine, castor oil or derivatives like Kolliphor EL (polyethoxylated castor oil, a nonionic surfactant) are also used as a drug delivery vehicle, e.g., for very nonpolar drugs such as the anti-cancer drugs paclitaxel and docetaxel (for references, see Patel et al. 2016). Furthermore, castor oil has been used as a solvent, co-solvent, stabilizing agent, and polyol for the formation of polymer-nanoparticle composites. It provides a facile route of nanoparticle surface functionalization and nanoparticle coating with polymers (Mensah et al. 2018).

Ricin as immunoconjugate/immune toxin in tumor therapy

Ricin has great therapeutic potential, for example, as an anti-cancer agent, in bone marrow transplantation, or in cell-based research, and is even explored in the context of nanoparticle formulations in tumor therapy.

The cytotoxic effects of ricin on tumor cells were already observed in the early 1950s, and thus well before the description of ricin's molecular structure and mode of action (Mosinger 1951). Initial findings in sarcomas in rat were further extended by Lin and co-worker who described prolonged survival of mice bearing Ehrlich ascites tumors upon ricin treatment (Lin et al. 1973), and in a human xenograft mouse model (Fodstad and Olsnes 1977). The observed higher sensitivity of cancer cells towards ricin may be attributed to their higher proliferation rate, requiring a higher rate of protein synthesis and thus making them more prone to ricin-mediated inhibition of this process, and/or to a higher ricin (B chain)-receptor concentration on the surface of tumor cells, allowing for enhanced uptake. An initial phase I study in 54 cancer patients with advanced disease indicated some therapeutic effects in a few patients (Fodstad et al. 1984). Still, however, this binding specificity was found insufficient, with ricin binding to virtually every cell type (Audi et al. 2005). Consequently, this led to the development of immunotoxins instead. In this approach, the biologically active component (here: ricin A chain) is covalently coupled to an antibody or other ligand that serves as binding moiety for selectively binding to the target cells (here: tumor cells). While initial ricin A-chain immunotoxins were found highly selective towards their target cells, their efficacy was poor (Neville Jr and Youle 1982; Wawrzynczak et al. 1991). This discrepancy was attributed to the mechanism of uptake and intracellular trafficking, with its processing in the endosomal/lysosomal system leading to rapid degradation, thus preventing the ricin A chain to reach the cytoplasm and thus its site of action (Preijers et al. 1988; van Horssen et al. 1995). In contrast, higher activity was achieved upon routing of immunotoxins through the Golgi complex (Press et al. 1986). A shift towards trafficking through the Golgi complex also provided the explanation for the finding of enhanced toxicity of a ricin A-chain immunotoxin upon addition of free ricin B chain (Neville Jr and Youle 1982; Timar et al. 1991). In addition to its binding properties, the ricin B chain was also found to have a role in the translocation of the membrane-bound A chain to the cytoplasm and in its intracellular routing (Timar et al. 1991), and to exert some pro-apoptotic effects by itself (Hasegawa et al. 2000). Consequently, immunotoxins based on the conjugation of the whole ricin molecule, rather than its A chain alone, were found to be more efficient (Olsnes 2004 and references therein). In this context, it should also be noted that, beyond its inhibitory effect on protein synthesis, other modes of action

have been described. These include the release of pro-inflammatory cytokines, lipid bilayer destabilization, and the direct induction of apoptosis (Griffiths et al. 1987; Hughes et al. 1996; see also Suppl. Fig. 4). The latter is mainly, but perhaps not exclusively, triggered by the intrinsic pathway (activation of caspases 9 and 3), increased ROS (reactive oxygen species) production, and DNA fragmentation/impaired DNA repair (Komatsu et al. 1998; Kumar et al. 2003; Wu et al. 2004; Rao et al. 2005), leading to various downstream effects (Slominska-Wojewodzka and Sandvig 2013 and references therein).

Initially, the ricin A chain, devoid of its B chain, was directly coupled to antibodies. This approach, however, could not prevent non-specific binding and uptake of the A chain (even without its B-chain-based binding properties) to various cells including macrophages and Kupffer cells (Fulton et al. 1988). This was mediated by carbohydrate residues on the A chain serving as binding partners for mannose receptors on the liver cells (Bourrie et al. 1986). Consequently, side effects were reduced and lifetimes in mice were prolonged by chemical deglycosylation of the A chain (Blakey et al. 1987; Fulton et al. 1988). Beyond the above-mentioned roles of the B chain leading to enhanced A-chain cytotoxicity, a protective role against cathepsin-mediated proteolysis of the A chain was described as well (Bilge et al. 1994). Further improvement was achieved by generating an altered ricin, the so-called blocked ricin, based on chemically blocking two galactose-binding sites of ricin by its chemical modification with affinity ligands (Lambert et al. 1991). Despite substantially reduced binding activity and cytotoxicity of blocked ricin, the catalytic activity of the A chain and the translocation role of the B chain were retained (Lambert et al. 1991).

Over the last decades, considerable work has been published on immunotoxins and other toxin conjugates. This also includes ricin, and various preclinical and clinical studies have been conducted (see Table 5 and Slominska-Wojewodzka and Sandvig 2013; Tyagi et al. 2015).

Major side effects associated with ricin-based immunotoxins are its low blood circulation time, immunogenicity, non-specific binding, and, as main side effect of ricin-derived immunotoxins, the vascular leak syndrome (VLS). Ribosome-inactivating proteins are often associated with high immunogenicity, thus limiting efficacy of treatment and the time period until an antibody response is detected, possibly leading to severe side effects. VLS, induced by ricin reaching the capillary endothelial cells, is a common dose-limiting toxicity and is associated with tissue edema and multi-organ failure (Baluna et al. 1999). It involves vascular endothelial cell damage, extravasation of fluids and proteins into tissues, interstitial edema, and organ failure (Baluna and Vitetta 1997). Despite detailed knowledge on the mechanism, it is believed to be mediated by the A chain rather than the cell-binding B chain.

Ricin in nanoparticle formations

In the light of unwanted side effects associated with the potential use of ricin in tumor therapy, several studies have looked into various nanoparticle formulations of ricin or the ricin A chain. In general, nanoparticles for the transport of drugs are explored with regard to altering biodistribution towards preferential delivery into on-target organs over off-target organs, enhanced blood circulation time/reduced degradation and elimination, increased penetration through biological and physiological barriers, enhanced efficacy of cellular uptake, and improved intracellular trafficking, for example, with regard to endosomal release. When decorating nanoparticles with specific ligands, efficient and selective targeting of the cells of interest is feasible as well. Altogether, these properties aim at leading to improved bioavailability and/or reduced side effects in non-target organs/cells. Lipid-based nanoparticles are the most advanced carriers, and have been extensively explored for ricin delivery. Inorganic nanoparticles may offer advantages with regard to stability and non-sensitivity against enzymatic or microbial degradation, but this non-biodegradability can also be an issue. Polymeric systems have gained increasing importance for example in the field of oligonucleotide delivery, and allow for numerous possibilities for structure modifications and custom-designable properties.

In oncology, nanoparticle delivery can benefit from the so-called passive tumor targeting due to the enhanced permeability and retention (EPR) effect, based on blood vessels presenting large fenestrations and gaps, distinct vessel irregularities, increased permeability, and lack of a normal lymph drainage as a consequence of increased angiogenesis and hypervascularization (Jang et al. 2003; Pecot et al. 2011). Thus, after transfer through endothelial fenestrations (permeability) facilitated by their nanosize, nanoparticles show a tendency to accumulate in the tumor tissue and are retained in the irregularly structured tissue (retention). Beyond this, active targeting explores the selective accumulation of nanoparticles on the surface of cancer cells, based on the binding of selective ligands presented on the nanoparticle surface.

As early as in the late 1970s, the encapsulation of ricin into negatively charged liposomes was described, leading to enhanced cytotoxicity or the circumvention of cellular ricin resistance (Nicolson and Poste 1978; Dimitriadis and Butters 1979). The latter was also true upon entrapping ricin into unilamellar liposomes (Gardas and Macpherson 1979). A considerable number of studies showed that monesin, a carboxylic ionophore and lysosomotropic agent, was able to potentiate the activity of ricin or ricin-based immunotoxins. This was true for monesin encapsulated into various liposomes, stealth immunoliposomes, or polymeric nanoparticles as well as for the free compound (Madan and Ghosh 1992; Vasandani et al. 1992; Griffin et al. 1993; Madan et al. 1993; Singh et al. 1994;

Table 5 Overview of clinical studies on ricin immunotoxins (for details on the clinical trials, see the references given)

Tumor entity	Binding partner	Study type	Observed therapeutic effects	Reference
Metastatic malignant melanoma	Anti-melanoma	Phase I	Treatment well tolerated with transient side effects; results suggested biological activity	Spitler et al. (1987)
Metastatic colon cancer	Antibody recognizing an antigen on the surface of CRC cells	Phase I	5/17 patients with biological activity as mixed tumor regression	Byers et al. (1989)
Non-Hodgkin's lymphoma	Anti-CD22	Phase I	PR (38%)	Vitetta et al. (1991)
Cutaneous T cell lymphoma	Anti-CD5	Phase I	PR (4/14)	LeMaistre et al. (1991)
Metastatic malignant melanoma	Anti-melanoma	Phase I/II	Evaluation of the role of cyclosporine A in blocking the antibody response; PR (1/9), SD (1/9)	Selvaggi et al. (1993)
Non-Hodgkin's lymphoma; children with ALL	Anti-CD22 (murine)	Phase I	Major responses in up to 30% of patients; dose limitation due to VLS	Amlot et al. (1993), Sausville et al. (1995)
Relapsed and refractory B cell neoplasms (NHL, CLL, ALL)	Anti-CD19	Phase I	CR (2/34), PR (3/34), transient responses (11/34)	Grossbard et al. (1993)
Metastatic colon cancer	Antibody recognizing an antigen on the surface of CRC cells	Phase I	No antitumor activity	LoRusso et al. (1995)
Non-Hodgkin's lymphoma	Anti-CD19	Phase I	CR (1/23), PR (1/23) in the bolus schedule group; PR (1/9) in the continuous-infusion group	Stone et al. (1996)
Hodgkin's lymphoma, CD25-positive	Anti-CD25	Phase I	PR (2/15)	Engert et al. (1997)
T cell lymphoma	Anti-CD7	Phase I	PR (2/11)	Frankel et al. (1997)
Leptomeningeal spread of systemic neoplasia	Anti-TfR	Pilot study	> 50% reduction of tumor cell counts in the lumbar CSF	Laske et al. (1997)
SCLC	Anti-CD56	Phase I	PR (1/21)	Lynch Jr. et al. (1997)
Non-Hodgkin's lymphoma	Cocktail of anti-CD22 + CD19	Phase I	PR in only 2/22 patients, perhaps due to HD37 aggregate formation	Messmann et al. (2000)
SCLC	Anti-CD56	Phase II	1 fatal incident of CLS and a nearly universal development of human anti-mouse and anti-ricin antibodies	Fidias et al. (2002)
Hodgkin's lymphoma, CD30 positive	Anti-CD30	Phase I	PR (1/15)	Schnell et al. (2002)
Hodgkin's lymphoma, CD25/CD30 positive	Anti-CD25/anti-CD30	Phase I/II	PR (2/27)/PR (1/15)	Schnell et al. (2003)
Children with ALL	Cocktail of anti-CD22 + CD19	Phase I	CR (3/17); decrease of > 95% in their peripheral blood blast counts (6/17); decrease of 75% (1/17)	Herrera et al. (2009)
Refractory or relapsed B-lineage-ALL	Cocktail of anti-CD22 + CD19	Phase I	PR (1/17); decreased blast counts (17/17)	Schindler et al. (2011)
B cell lymphoma, CD19 positive	Anti-CD19	Phase III	No effect in consolidative therapy after bone marrow transplant	Furman et al. (2011)
Multiple bladder cancer	BCMab1 (against aberrantly glycosylated integrin $\alpha 3 \beta 1$)	Pilot study in 1 patient	Complete remission	Li et al. (2017)

Abbreviations: *ALL* acute lymphoblastic leukemia, *CLL* chronic lymphocytic leukemia, *CLS* capillary leak syndrome, *CR* complete remission, *CSF* cerebrospinal fluid, *NHL* non-Hodgkin's lymphoma, *PR* partial response, *SCLC* small-cell lung cancer, *SD* stable disease, *TfR* transferrin receptor, *VLS* vascular lead syndrome

Ferdous et al. 1998; Shaik et al. 2001; Singh et al. 2001; Tyagi et al. 2013). Notably, this effect was observed not only in the case of free ricin, but also for liposomal formulations, indicating endocytotic uptake of liposomal ricin (Bharadwaj et al. 2006). When comparing different liposomal formulations, major differences were found, with ricin cytotoxicity being dependent on the surface charge and the degree of polyethylene glycol (PEG) substitution, and certain negatively charged liposomes offering maximum cytotoxicity (Rathore and Ghosh 2008; Tyagi et al. 2011; Tyagi et al. 2015). Interestingly, a similar approach of liposome formulation has also been explored for intracellular delivery of anti-ricin A-chain monoclonal antibodies to neutralize ricin toxicity, indicating the potential utility of cell-permeable antibodies for post-exposure treatment of ricin intoxication (Wu et al. 2010). Beyond describing *in vitro* effects in cell culture, several studies also explored the *in vivo* use of ricin formulations in mice, demonstrating enhanced cytotoxicity and reduced toxic side effects (Vasandani et al. 1992; Madan et al. 1993; Zhang et al. 1999). For example, prolonged survival was observed in an H-MESO-1 tumor xenograft model, or, in a subset of the treated mice, even the absence of intraperitoneal tumors (Griffin et al. 1993).

Finally, targeted delivery has been explored in the context of ricin as well. While a few papers have looked into the use of the ricin B chain as binding moiety for quantum dot, liposome, or carbon dot targeting (Friede and von Holt 1991; Iversen et al. 2012; Li et al. 2018), most studies have aimed at further improving ricin cytotoxicity and reducing side effects due to more selective cellular uptake. Liposomal ricin with 0.5% Folate-PEG for folate receptor-mediated targeted delivery (Tyagi and Ghosh 2011), peptide-targeted mesoporous silica nanoparticle-supported lipid bilayers (“protocells”) for peptide-directed, cell-specific uptake (Epler et al. 2012), multiwalled carbon nanotubes (MWNTs) for selective delivery of ricin to mamma carcinoma cells overexpressing human epidermal growth factor receptor 2 (HER2/neu; Weng et al. 2009) or virus-like particles (VLPs) of bacteriophage MS2, modified with a peptide (SP4) and a histidine-rich fusogenic peptide (H5WYG) for promoting endosomal escape (Ashley et al. 2011), have been described. These studies followed up on a very early publication on ricin or the ricin A chain encapsulated in liposomes bearing an anti-carcinoembryonic antigen (CEA) antibody, for selective delivery into CEA-expressing tumor cells (Watanabe and Osawa 1987). Finally, a most recent study engineered CXCR4-targeted nanoparticles via self-assembly of the modular protein T22-mRTA-H6, a recombinant version of ricin. Beyond high cytotoxicity in CXCR4+ tumor cells *in vitro*, profound therapeutic anti-tumor activity in the absence of toxic side effects was also observed in a mouse model of acute myeloid leukemia (Diaz et al. 2018).

Ricin as weapon

As described above, ricin is one of the most potent and lethal biological substances known and is thus also considered as important bioweapon. The broad distribution of the native plant, the broad availability of castor beans as well as the easy process of isolation/extraction of the toxin and its increasing popularity through the Internet make ricin nowadays to an attractive agent in the context of bioterroristic activities (Poelchen and Wirkner 2003; Olsnes 2004; Roxas-Duncan and Smith 2012; Thiermann et al. 2013; Robert Koch Institut 2017).

During World War I and II, the USA and other countries investigated ricin for its military potential as a biological weapon (e.g., bullets and shrapnels coated with ricin, or as “dust cloud”). Ricin is known as “Agent W” (compound W, because of the military symbol W for ricin) (see Olsnes 2004; Roxas-Duncan and Smith 2012; Diac et al. 2017; Patocka 2018 and references therein). According to some reports, ricin has possibly been used as a warfare agent in Iraq in the 1980s and more recently by terrorist organizations (<https://emergency.cdc.gov/agent/ricin/facts.asp>).

Nowadays, ricin is listed (i) in the Biological Weapons Convention (1972) and (ii) in the Chemical Weapons Convention (1997). More specifically, (i) based on its profound lethality and high availability, ricin has been categorized as a category B biothreat agent (second-highest priority) by the United States (US) Centers for Disease Control and Prevention (CDC). (ii) Ricin is also among the toxic chemicals listed in schedule 1 of the Chemical Weapons Convention (CWC), and in the Biological and Toxin Weapons Convention (BTWC). Its possession, transfer, purification, and use are subject to domestic and international regulations, and are controlled by the Organization for the Prohibition of Chemical Weapons (OPCW) (for references, see Poelchen and Wirkner 2003; Worbs et al. 2011; Roxas-Duncan and Smith 2012; Arnold 2017; <https://www.opcw.org/our-work/what-chemical-weapon>).

Ricin antibodies and immunization for protection and therapy

There is ongoing research on treatment options after ricin intoxication as well as on potential vaccine candidates (Slominska-Wojewodzka and Sandvig 2013; Hu et al. 2014; Respaud et al. 2016; Brey et al. 2016; Noy-Porat et al. 2017; Rosenfeld et al. 2017; Toth et al. 2017 and references therein).

Therapeutic strategies after ricin intoxication may include disease-modifying countermeasures, based on anti-ricin antibodies, small molecule compounds, and various combinations of those (immunomodulators and other pharmacological-

based treatment options; see Roxas-Duncan and Smith 2012; Hu et al. 2014; Gal et al. 2017). Passive protection with aerosolized anti-ricin immunoglobulin (IgG) has been evaluated as prophylaxis prior to aerosol challenge as well (see Roxas-Duncan and Smith 2012). Neutralizing antibodies are the most promising post-exposure treatment of ricin intoxication, yet so far they have been shown to be effective only when given within several hours post exposure (Noy-Porat et al. 2017). Various monoclonal antibodies have been generated (for review, see Hu et al. 2014), and it has been shown that the use of antibody combinations leads to improved neutralizing capacity in vitro, in vivo and in pre-clinical studies (Prigent et al. 2011). A tri-antibody based cocktail showed high efficacy and very high survival rates (> 70%) when animals were treated as late as 48 h post exposure, and significant protection (> 30%) even at 72 h (Noy-Porat et al. 2017).

A recombinant ribotoxic A-chain subunit with mutations in two residues, named RiVax, has been developed as a specific vaccine against ricin (Smallshaw et al. 2005; Vitetta et al. 2006). RiVax is safe in rabbits and mice, induces high titers of neutralizing antibodies when administered i.m., and had sufficient pre-clinical safety data (Smallshaw et al. 2005). Results from a phase I human trial indicated that RiVax elicited ricin-neutralizing antibodies and was well tolerated in humans (Vitetta et al. 2005; Vitetta 2006). Two closely related RTA-based subunit vaccines, RiVax (see above) and RVEc (a truncated derivative of RTA), are now under further development (for review, see Brey III et al. 2016).

Another potent anti-ricin neutralizing, humanized antibody (hD9) exhibited high efficacy in a female BALB/c mouse model. More specifically, a dose of 5 µg hD9 per mouse (0.25 mg/kg) could rescue 100% of the mice, e.g., after $5 \times \text{LD}_{50}$ (50 µg/kg) ricin challenge (Hu et al. 2014 and references therein).

Gal and co-workers reviewed treatment options upon pulmonary ricin intoxication (Gal et al. 2017). They discuss (i) small-molecule compounds and their potential therapeutic benefit against ricinosis by directly interfering with the toxin or influencing ricin's intracellular trafficking, as well as (ii) the co-administration of anti-ricin antibodies with immunomodulatory drugs for attenuating lung injury via neutralizing the toxin.

Ricin detection

For rapid detection of ricin in environmental and clinical samples as well as in food, analytical methods with high selectivity and sensitivity are required (Kalb and Barr 2009). It is hereby of great importance to distinguish between functionally active vs. denatured ricin, since this has important implications on emergency operating schedules, therapeutic measures, and forensic analysis. This relies on performing functional assays, which also allow for the differentiation between A-chain vs. B-chain activity, or both (see Duracova et al. 2018).

With respect to forensic analysis, ricin detection in clinical samples is particularly difficult. The fact that ricin is denatured by temperatures over 80 °C is important in the context of its analysis, e.g., in food. Furthermore, as mentioned above, agglutinin as well as several toxic proteins and isoforms have been purified from the seeds of *Ricinus communis*, with amino acid compositions of the isoforms being very similar. Consequently, considerable research efforts have been focused on the development of sensitive methods for the detection of ricin D in complex matrices such as human biological samples, environmental samples, water, and food (Kalb and Barr 2009; Severino et al. 2012; Worbs et al. 2015; Stern et al. 2018; Weber 2018). Possibilities of ricin detection are the lateral flow assay (LFA), the detection of ricinin as accompanying alkaloid in castor bean seeds, polymerase chain reaction (PCR)-based detection of DNA (only applicable in castor bean seeds), or liquid chromatography/mass spectrometry (LC/MS) after protein digestion (Weber and Schulz 2011; Worbs et al. 2015; Duracova et al. 2018; Weber 2018). Furthermore, a variety of immunological methods for ricin detection exist, whereby enzyme-linked immunosorbent assays (ELISA) has been proven to be one of the most versatile techniques (for review, see Severino et al. 2012).

(i) A rapid test based on the immunological detection of ricin in beverages, food, and consumer products is described by Weber and Schulz (2011). The unmodified toxin can be detected with sufficient sensitivity by LFA or ELISA (Weber 2014). When metabolized, ricin is no longer detectable in the body. This also means that, when ricin intoxication is suspected, the environment of the affected person should be analyzed retrospectively with regard to possible sources of ricin uptake. Using LFA, the detection limit is 0.05 mg/L in water and 1–2.5 mg/kg in food. A forensic validation procedure can be performed by peptide analysis (Weber and Schulz 2011).

(ii) For the unambiguous identification of ricin toxin (and the alkaloid marker ricinine) from crude plant materials, special LC/MS and matrix-assisted laser desorption/ionization time-of-flight (MALDI-TOF) MS methods have been developed (Darby et al. 2001). Nowadays, the combination of different techniques is used (Worbs et al. 2015; Stern et al. 2018). Additionally, a ricin forensic profiling approach based on a complex set of biomarkers including carbohydrates, fatty acids, seed storage proteins in combination with data on ricin, and *Ricinus communis* agglutinin is developed (Fredriksson et al. 2018).

In samples with complex matrices, MS assays have been found capable of unambiguously identifying ricin and its activity (Kalb et al. 2015). More specifically, Kalb and co-workers reported on the application of MS-based methods for detection, differentiation, and quantitation of ricin and RCA120 in blinded samples, as part of the EQuATox proficiency test (Establishment of Quality Assurance for the Detection of Biological Toxins of Potential Bioterrorism Risk; Kalb et al. 2015). This was the first program for the

investigation of qualitative and quantitative data on an international level, aiming at the establishment of an assortment of ricin detection methods which employ well-characterized analytical standards (Kalb et al. 2015).

(iii) Ricinine is used as surrogate marker for ricin exposure and may be more robust than the detection of ricin itself. Due to its chemical stability and extractability in routine extraction procedures in toxicology laboratories, ricinine can be identified by MS-based untargeted toxicology screening (Weber 2014; Lopez Nunez et al. 2017). Ricinin is heat stable and not rapidly metabolized (Johnson et al. 2005; Weber 2014; Weber 2018). Verougstraete and co-workers described an LC-MS/MS method for the detection of ricinine in serum, blood, and urine, validated according to European Medicines Agency (EMA) guidelines and successfully applied to patient samples (Verougstraete et al. 2018). Of note, however, ricinine may also be detectable in the general population since many consumer products contain castor oil. This has been shown, for example, in a study of Pittman and co-workers who characterized urinary ricinine concentrations from 989 individuals presumably unexposed to ricin (Pittman et al. 2013).

***Ricinus communis*-an interesting and unusual plant**

In the light of the high toxicity of the components in the *Ricinus communis* plant, the “mysterious” ricinus-feeding worm (in the book Jonah, Bible; see also the “Introduction”) has been of special scientific interest considering its resistance. Nowadays, the Israeli tiger moth, *Olepa schleinii* (Lepidoptera, Arctiidae), which regularly infests *Ricinus communis* fits perfectly to the detailed description of this ricinus feeder mentioned in the Bible (Hausmann and Müller 2006). The leaves of the ricinus plant are also the preferential feed of the Asian population of the silkworm (*Samia cynthia ricini*) in India (Vijayan et al. 2006). These examples identify *Ricinus communis* as host and food plant for insects including moths and butterflies as well as some lepidopteran larvae and birds (Roxas-Duncan and Smith 2012), despite its superb toxicity on other larvae of parasites, aphids, gnats, and others. However, the latter aspect is of particular interest with regard to increasing concerns of pesticide accumulation in the environment, prompting researchers to develop safer alternatives. Indeed, some insecticidal lectins may be useful in contributing to the development of integrated pest management strategies with minimal effect(s) on non-target organisms. Plant-derived, biodegradable materials are currently evaluated as an alternative remedy in controlling arthropods of medical and veterinary relevance (Singh and Kaur 2017).

Castor oil, as described above, offers a number of interesting applications and is considered as an option for biodiesel production in several countries. Its importance

as a bioenergy and industrial feedstock results from the potential of modifications in fatty acid composition, very high oil yields, a wide range of adaptation, and the plant's ability to be grown on marginal sites subject to drought and saline conditions (Severino et al. 2012). A life-cycle analysis revealed that the use of castor oil for biodiesel production could offer many advantages like (i) an energy return-on-energy investment (EROEI) of 2.60 and (ii) a positive contribution to climate-change reduction, as revealed by a positive carbon balance (Amouri et al. 2017). In this context, the development of low-ricin or ricin-free castor cultivars (as well as low-ricin, low-ricinine, low-allergen cultivars) is of special interest for commercial castor oil production (Severino et al. 2012). Sousa and co-workers explored the concept of gene knockdown by RNA interference (RNAi) in order to silence the ricin-encoding genes in the endosperm of castor beans, aiming at increased safety of castor bean cultivation for farmers, industrial workers, and society (Sousa et al. 2017). Furthermore, after oil extraction, bio-detoxified castor bean cake is attractive for animal feeding due to its rich content in valuable proteins (Sousa et al. 2017). In contrast, however, since lectins such as ricin play an important role in plant protection against insect pests, the development of ricin-free castor genotypes may increase the risk of pest incidence rates (for review, see Vandenborre et al. 2011).

The need to generate larger and highly standardized amounts of ricin and its modified forms for possible pharmacological applications has led to the development of strategies based on genetic engineering, e.g., the expression of the two full-length polypeptide chains in transgenic tobacco (Sehnke et al. 1994; Sehnke and Ferl 1999). It is interesting to note that the emerging field of nanomedicines, allowing for improved pharmacokinetics and putatively leading to targeted drug delivery, may help in controlling unwanted side effects.

Thus, ricin is perhaps one of the most impressive examples of compounds with superb toxicity on the one hand and therapeutic utility on the other.

Acknowledgments Parts of this article on the history of the plant *Ricinus communis* in the Papyrus Ebers were presented at the Symposium on the Occasion of the 30. Anniversary of the Postgraduate Study Program (PGS) “Toxicology and Environmental Protection” at the University of Leipzig (March 23, 2018). PGS graduation theses by Jörg Pietsch (2004), Gabriele Baranius (2009), and Thorsten Meißner (2017) as well as a study work prepared by Avina Graefe (2012) contributed to this review article and are greatly acknowledged. The authors are grateful to Dr. Jens Grosche (Effigis AG) for providing Fig. 4. The authors also wish to thank Adelgunde Graefe (PGS “Toxicology and Environmental Protection,” University of Leipzig) and Thomas Gruner (Library of the Medical Faculty, University of Leipzig) for their support.

Author contribution statement HF, RS, and AA wrote the manuscript. HF took the photos, HF and AA created figures and tables. All authors read and approved the manuscript.

Compliance with ethical standards

Not applicable.

Conflict of interest The authors declare that they have no conflicts of interest.

References

- Abal P, Louzao MC, Antelo A, Alvarez M, Cagide E, Vilariño N, Vieytes MR, Botana LM (2017) Acute oral toxicity of tetrodotoxin in mice: determination of lethal dose 50 (LD50) and no observed adverse effect level (NOAEL). *Toxins (Basel)* 9(3):pii: E75. <https://doi.org/10.3390/toxins9030075>
- Abbas M, Ali A, Arshad M, Atta A, Mehmood Z, Tahir IM, Iqbal M (2018) Mutagenicity, cytotoxic and antioxidant activities of *Ricinus communis* different parts. *Chem Cent J* 12(1):3. <https://doi.org/10.1186/s13065-018-0370-0>
- Abdul WM, Hajrah NH, Sabir JSM, Al-Garni SM, Sabir MJ, Kabli SA, Saini KS, Bora RS (2018) Therapeutic role of *Ricinus communis* L. and its bioactive compounds in disease prevention and treatment. *Asian Pac J Trop Med* 11(3):177–185
- Amlot PL, Stone MJ, Cunningham D, Fay J, Newman J, Collins R, May R, McCarthy M, Richardson J, Ghetie V et al (1993) A phase I study of an anti-CD22-deglycosylated ricin A chain immunotoxin in the treatment of B-cell lymphomas resistant to conventional therapy. *Blood* 82:2624–2633
- Amouri M, Mohellebi F, Zaid TA, Aziza M (2017) Sustainability assessment of *Ricinus communis* biodiesel using LCA approach. *Clean Techn Environ Policy* 19:749–760. <https://doi.org/10.1007/s10098-016-1262-4>
- Araki T, Funatsu G (1987) The complete amino acid sequence of the B-chain of ricin E isolated from small-grain castor bean seeds. Ricin E is a gene recombination product of ricin D and *Ricinus communis* agglutinin. *Biochim Biophys Acta* 911:191–200
- Arnold W (2017) Rizinus, Wunderbaum - *Ricinus communis*: https://www.awl.ch/heilpflanzen/ricinus_communis/rizinus.htm. Accessed 18 March 2019
- Ashley CE, Carnes EC, Phillips GK, Durfee PN, Buley MD, Lino CA, Padilla DP, Phillips B, Carter MB, Willman CL, Brinker CJ, Caldeira Jdo C, Chackerian B, Wharton W, Peabody DS (2011) Cell-specific delivery of diverse cargos by bacteriophage MS2 virus-like particles. *ACS Nano* 5:5729–5745
- Audi J, Belson M, Patel M, Schier J, Osterloh J (2005) Ricin poisoning: a comprehensive review. *JAMA* 294:2342–2351. <https://doi.org/10.1001/jama.294.18.2342> PMID 16278363
- Balint GA (1974) Ricin: the toxic protein of castor oil seeds. *Toxicology* 2:77–102
- Baluna R, Vitetta ES (1997) Vascular leak syndrome: a side effect of immunotherapy. *Immunopharmacology* 37:117–132
- Baluna R, Rizo J, Gordon BE, Ghetie V, Vitetta ES (1999) Evidence for a structural motif in toxins and interleukin-2 that may be responsible for binding to endothelial cells and initiating vascular leak syndrome. *Proc Natl Acad Sci U S A* 96:3957–3962
- Bharadwaj S, Rathore SS, Ghosh PC (2006) Enhancement of the cytotoxicity of liposomal ricin by the carboxylic ionophore monensin and the lysosomotropic amine NH4Cl in Chinese hamster ovary cells. *Int J Toxicol* 25:349–359
- Bigi MFMA, Torkomian VLV, de Groote STCS, Hebling MJB, Bueno OC, Pagnocca FC, Fernandes JB, Vieira PC, da Silva MFGF (2004) Activity of *Ricinus communis* (Euphorbiaceae) and ricinine against the leaf-cutting ant *Atta sexdens rubropilosa* (hymenoptera: Formicidae) and the symbiotic fungus *Leucoagaricus gongylophorus*. *Pest Manag Sci* 60:933–938
- Bilge A, Howell-Clark J, Ramakrishnan S, Press OW (1994) Degradation of ricin A chain by endosomal and lysosomal enzymes—the protective role of ricin B chain. *Ther Immunol* 1:197–204
- Blakey DC, Watson GJ, Knowles PP, Thorpe PE (1987) Effect of chemical deglycosylation of ricin A chain on the *in vivo* fate and cytotoxic activity of an immunotoxin composed of ricin A chain and anti-Thy 1.1 antibody. *Cancer Res* 47:947–952
- Bourrie BJ, Casellas P, Blythman HE, Jansen FK (1986) Study of the plasma clearance of antibody–ricin-A-chain immunotoxins. Evidence for specific recognition sites on the A chain that mediate rapid clearance of the immunotoxin. *Eur J Biochem* 155:1–10
- Bradberry SM, Dickers KJ, Rice P, Griffiths GD, Vale JA (2003) Ricin poisoning. *Toxicol Rev* 22(1):65–70
- Brey RN III, Mantis NJ, Pincus SH, Vitetta ES, Smith LA, Roy CJ (2016) Recent advances in the development of vaccines against ricin. *Hum Vaccin Immunother* 5:1196–1201. <https://doi.org/10.1080/21645515.2015.1124202>
- Buch C (2018) Pflanzenporträts 2018; *Ricinus communis* - Rizinus, Wunderbaum (Euphorbiaceae), p 1–7, Bochumer Botanischer Verein, www.botanik-bochum.de. Accessed 18 March 2019
- Buonocore C, Alipour M, Omri A, Pucaj K, Smith MG, Suntres ZE (2011) Treatment of ricin A-chain-induced hepatotoxicity with liposome-encapsulated N-acetylcysteine. *J Drug Target* 1:1–9. ISSN 1061-186X print/ISSN 1029-2330 online. <https://doi.org/10.3109/1061186X.2011.582645>
- Burrows WD, Renner SE (1999) Biological warfare agents as threats to potable water. *Environ Health Perspect* 107:975–984
- Butterworth AG, Lord JM (1983) Ricin and *Ricinus communis* agglutinin subunits are all derived from a single-size polypeptide precursor. *Eur J Biochem* 137:57–65. <https://doi.org/10.1111/j.1432-1033.1983.tb07795.x>
- Byers VS, Rodvien R, Grant K, Durrant LG, Hudson KH, Baldwin RW, Scannon PJ (1989) Phase I study of monoclonal antibody–ricin A chain immunotoxin XomaZyme-791 in patients with metastatic colon cancer. *Cancer Res* 49:6153–6160
- Cazal C d M, Batalhão JR, Domingues Vde C, Bueno OC, Filho ER, Forim MR, da Silva MF, Vieira PC, Fernandes JB (2009) High-speed counter-current chromatographic isolation of ricinine, an insecticide from *Ricinus communis*. *J Chromatogr A* 1216(19):4290–4294. <https://doi.org/10.1016/j.chroma.2009.02.008>
- Challoner KR, McCarron MM (1990) Castor bean intoxication: review of reported cases. *Ann Emerg Med* 19:1177–1183
- Chen Z, Zhang J, Chen G (2008) Simultaneous determination of flavones and phenolic acids in the leaves of *Ricinus communis* Linn. By capillary electrophoresis with amperometric detection. *J Chromatogr B Analyt Technol Biomed Life Sci* 863(1):101–106. <https://doi.org/10.1016/j.jchromb.2008.01.002>
- Cook DL, David J, Griffiths GD (2006) Retrospective identification of ricin in animal tissues following administration by pulmonary and oral routes. *Toxicology* 223:61–70
- Crompton R, Gall D (1980) Georgi Markov - death in a pellet. *Medico-Legal J* 48:51–62
- Dai J, Zhao L, Yang H, Guo H, Fan K, Wang H, Qian W, Zhang D, Li B, Wang H et al (2011) Identification of a novel functional domain of ricin responsible for its potent toxicity. *J Biol Chem* 286:12166–12171
- Darby SM, Miller ML, Allen RO (2001) Forensic determination of ricin and the alkaloid marker ricinine from castor bean extracts. *J Forensic Sci* 46(5):1033–1042 PMID: 11569541
- de la Cruz R, Pastor AM, Delgado-García JM (1994) The neurotoxic effects of *Ricinus communis* agglutinin-II. *J Toxicol Toxin Rev* 14(1):1–46. <https://doi.org/10.3109/155695949509089967>
- Despeyroux D, Walker N, Pearce M, Fisher M, McDonnell M, Bailey SC, Griffiths GD, Watts P (2000) Characterization of ricin heterogeneity

- by electrospray mass spectrometry, capillary electrophoresis, and resonant mirror. *Anal Biochem* 279:23–36
- Deus-de-Oliveira N, Felix SP, Carriello-Gama C, Fernandes KV, Damatta RA, Machado OL (2011) Identification of critical amino acids in the IgE epitopes of Ric c 1 and Ric c 3 and the application of glutamic acid as an IgE blocker. *PLoS One* 6(6):e21455. <https://doi.org/10.1371/journal.pone.0021455>
- Diac M, Matei MC, Manea C, Schiopu C, Iliescu DB, Furnica C, Chistol RO, Knieling A (2017) Intoxication with ricin-biochemical weapon. *Rev Chim* 68(6):1329–1332
- Diaz R, Pallares V, Cano-Garrido O, Serna N, Sanchez-Garcia L, Falgas A, Pesarrodona M, Unzueta U, Sanchez-Chardi A, Sanchez JM, Casanova I, Vazquez E, Mangues R, Villaverde A (2018) Selective CXCR4(+) cancer cell targeting and potent antineoplastic effect by a nanostructured version of recombinant ricin. *Small* 14:e1800665
- Dimitriadis GJ, Butters TD (1979) Liposome-mediated ricin toxicity in ricin-resistant cells. *FEBS Lett* 98:33–36
- Duracova M, Klimentova J, Fucikova A, Dresler J (2018) Proteomic methods of detection and quantification of protein toxins. *Toxins* 10:99. doi:<https://doi.org/10.3390/toxins10030099>
- Eastmond PJ (2004) Cloning and characterization of the acid lipase from castor beans. *J Biol Chem* 279(44):45540–45545
- Elimam AM, Elmalik KH, Ali FS (2009) Larvicidal, adult emergence inhibition and oviposition deterrent effects of foliage extract from *Ricinus communis* L against *Anopheles arabiensis* and *Culex quinquefasciatus* in Sudan. *Trop Biomed* 26(2):130–139
- Endo Y, Tsurugi K (1987) RNA N-glycosidase activity of ricin A-chain. Mechanism of action of the toxic lectin ricin on eukaryotic ribosomes. *J Biol Chem* 262:8128–8130
- Endo Y, Mitsui K, Motizuki M, Tsurugi K (1987) The mechanism of action of ricin and related toxic lectins on eukaryotic ribosomes. The site and the characteristics of the modification in 28S ribosomal RNA caused by the toxins. *J Biol Chem* 262:5908–5912
- Engert A, Diehl V, Schnell R, Radszuhn A, Hatwig MT, Drillich S, Schon G, Bohlen H, Tesch H, Hansmann ML, Barth S, Schindler J, Ghetie V, Uhr J, Vitetta E (1997) A phase-I study of an anti-CD25 ricin A-chain immunotoxin (RFT5-SMPT-dgA) in patients with refractory Hodgkin's lymphoma. *Blood* 89:403–410
- Epler K, Padilla D, Phillips G, Crowder P, Castillo R, Wilkinson D, Wilkinson B, Burgard C, Kalinich R, Townson J, Chackerian B, Willman C, Peabody D, Wharton W, Brinker CJ, Ashley C, Carnes E (2012) Delivery of ricin toxin a-chain by peptide-targeted mesoporous silica nanoparticle-supported lipid bilayers. *Adv Healthc Mater* 1:348–353
- Fabbrini MS, Katayama M, Nakase I, Vago R (2017) Plant ribosome-inactivating proteins: progresses, challenges and biotechnological applications (and a few digressions). *Toxins (Basel)* 9(10):pii: E314. <https://doi.org/10.3390/toxins9100314> PMID:29023422; PMCID:PMC5666361
- Ferdous AJ, Stemberge NY, Singh M (1998) Role of monensin PLGA polymer nanoparticles and liposomes as potentiators of ricin A immunotoxins *in vitro*. *J Control Release* 50:71–78
- Ferraz AC, Angelucci MEM, Da Costa ML, Batista IR, de Oliveira BH, da Cunha C (1999) Pharmacological evaluation of ricinine, a central nervous system stimulant isolated from *Ricinus communis*. *Pharmacol Biochem Behav* 63:367–375
- Ferraz AC, Anselmo-Franci JA, Perosa SR, de Castro-Neto EF, Bellissimo MI, de Oliveira BH, Cavalheiro EA, Naffah-Mazzacoratti MDG, da Cunha C (2002) Amino acid and monoamine alterations in the cerebral cortex and hippocampus of mice submitted to ricinine-induced seizures. *Pharmacol Biochem Behav* 72:779–786
- Fidias P, Grossbard M, Lynch TJ Jr (2002) A phase II study of the immunotoxin N901-blocked ricin in small-cell lung cancer. *Clin Lung Vanc* 3:219–222
- Fodstad O, Olsnes S (1977) Studies on the accessibility of ribosomes to inactivation by the toxic lectins abrin and ricin. *Eur J Biochem* 74:209–215
- Fodstad O, Olsnes S, Pihl A (1976) Toxicity, distribution and elimination of the cancerostatic lectins abrin and ricin after parenteral injection into mice. *Br J Cancer* 34:418–425
- Fodstad O, Kvalheim G, Godal A, Lotsberg J, Aamdal S, Host H, Pihl A (1984) Phase I study of the plant protein ricin. *Cancer Res* 44:862–865
- Foxwell BM, Detre SI, Donovan TA, Thorpe PE (1985) The use of anti-ricin antibodies to protect mice intoxicated with ricin. *Toxicology* 34:79–88
- Frankel AE, Laver JH, Willingham MC, Burns LJ, Kersey JH, Vallera DA (1997) Therapy of patients with T-cell lymphomas and leukemias using an anti-CD7 monoclonal antibody-ricin A chain immunotoxin. *Leuk Lymphoma* 26:287–298
- Franz DR (1997) Defense against toxin weapons. In: Sidell FR et al (eds) *Textbook of military medicine, part 1: medical aspects of chemical and biological warfare*. Office of the Surgeon General, Department of the Army, Washington D.C., pp 603–620
- Franz D, Jaax N (1997) Ricin toxin. In: Zajchuk R, Bellamy RF (eds) *Textbook of military medicine: part 1, medical aspects of chemical and biological warfare*. Office of the Surgeon General of the Army, TMM Publications, Washington, pp 631–642
- Fredriksson SÅ, Wunschel DS, Lindström SW, Nilsson C, Wahl K, Åstot C (2018) A ricin forensic profiling approach based on a complex set of biomarkers. *Talanta* 186:628–635. <https://doi.org/10.1016/j.talanta.2018.03.070>
- Friede MH, von Holt C (1991) Ricin B-chain promotes the internalisation of liposomal contents into rat hepatoma cells. *Biochim Biophys Acta* 1069:273–280
- Friese J, Gleitz J, Gutser UT, Heubach JF, Matthiesen T, Wilffert B, Selve N (1997) Aconitum sp. alkaloids: the modulation of voltage-dependent Na⁺ channels, toxicity and antinociceptive properties. *Eur J Pharmacol* 337(2–3):165–174
- Frohne D, Pfänder HJ (2004) *Giftpflanzen- Ein Handbuch für Apotheker, Ärzte, Toxikologen und Biologen*. 5. Auflage. Wissenschaftliche Verlagsgesellschaft mbH, Stuttgart
- Fulton RJ, Uhr JW, Vitetta ES (1988) *In vivo* therapy of the BCL1 tumor: effect of immunotoxin valency and deglycosylation of the ricin A chain. *Cancer Res* 48:2626–2631
- Funatsu G, Funatsu M (1977) Separation of the two constituent polypeptide chains of ricin D. *Agric Biol Chem* 41:1211–1215
- Furman RR, Grossbard ML, Johnson JL, Pecora AL, Cassileth PA, Jung SH, Peterson BA, Nadler LM, Freedman A, Bayer RL, Bartlett NL, Hurd DD, Cheson BD, Cancer Leukemia Group B, Eastern Cooperative Oncology G (2011) A phase III study of anti-B4-blocked ricin as adjuvant therapy post-autologous bone marrow transplant: CALGB 9254. *Leuk Lymphoma* 52:587–596
- Gage E, Hernandez MO, O'Hara JM, McCarthy EA, Mantis NJ (2011) Role of the mannose receptor (CD206) in innate immunity to ricin toxin. *Toxins* 3(9):1131–1145. <https://doi.org/10.3390/toxins3091131>
- Gal Y, Mazor O, Falach R, Sapoznikov A, Kronman C, Sabo T (2017) Treatments for pulmonary ricin intoxication: current aspects and future prospects. *Toxins* 9(10):E311. <https://doi.org/10.3390/toxins9100311>
- Garber EAE (2008) Toxicity and detection of ricin and abrin in beverages. *J Food Prot* 71:1875–1883
- Gardas A, Macpherson I (1979) Microinjection of ricin entrapped in unilamellar liposomes into a ricin-resistant mutant of baby hamster kidney cells. *Biochim Biophys Acta* 584:538–541
- Germer R (1982) Öle, In: *Lexikon der Ägyptologie IV, Megiddo-Pyramiden*, Hrsg. v. W Helck, Wiesbaden, Sp. 552
- Ghalioungui P (1987) *The Ebers Papyrus: a new English translation, commentaries, and glossaries*. Cairo

- Greenwood JS, Helm M, Gietl C (2005) Ricinosomes and endosperm transfer cell structure in programmed cell death of the nucellus during *Ricinus* seed development. *Proc Natl Acad Sci U S A* 102:2238–2243
- Griffin T, Rybak ME, Recht L, Singh M, Salimi A, Raso V (1993) Potentiation of antitumor immunotoxins by liposomal monensin. *J Natl Cancer Inst* 85:292–298
- Griffiths GD (2011) Understanding ricin from a defensive viewpoint. *Toxins* 3(11):1373–1392. <https://doi.org/10.3390/toxins3111373>
- Griffiths GD, Leek MD, Gee DJ (1987) The toxic plant proteins ricin and abrin induce apoptotic changes in mammalian lymphoid tissues and intestine. *J Pathol* 151:221–229
- Griffiths GD, Rice P, Allenby AC, Bailey C, Upshall DG (1995) Inhalation toxicology and histopathology of ricin and abrin toxins. *Inhal Toxicol* 7(2):269–288. <https://doi.org/10.3109/08958379509029098>
- Griffiths GD, Phillips GJ, Holley J (2007) Inhalation toxicology of ricin preparations: animal models, prophylactic and therapeutic approaches to protection. *Inhal Toxicol* 19:873–887
- Grossbard ML, Lambert JM, Goldmacher VS, Spector NL, Kinsella J, Eliseo L, Coral F, Taylor JA, Blattler WA, Epstein CL et al (1993) Anti-B4-blocked ricin: a phase I trial of 7-day continuous infusion in patients with B-cell neoplasms. *J Clin Oncol* 11:726–737
- Hänsel R, Keller K, Rimpler H, Schneider G (1994) Hagers Handbuch der Pharmazeutischen Praxis: Drogen P-Z Folgeband 2. Springer Verlag Berlin Heidelberg GmbH (Hager's handbook of pharmaceutical practice)
- Hartley MR, Lord JM (2004) Cytotoxic ribosome-inactivating lectins from plants. *Biochim Biophys Acta* 1701:1–14
- Hasegawa N, Kimura Y, Oda T, Komatsu N, Muramatsu T (2000) Isolated ricin B-chain-mediated apoptosis in U937 cells. *Biosci Biotechnol Biochem* 64:1422–1429
- Hassan Y, Ogg S, Ge H (2018) Expression of novel fusion antiviral proteins ricin a chain-pokeweed antiviral proteins (RTA-PAPs) in *Escherichia coli* and their inhibition of protein synthesis and of hepatitis B virus *in vitro*. *BMC Biotechnol* 18(1):47. <https://doi.org/10.1186/s12896-018-0458-6> PMID:30081895 PMCID: PMC6080542
- Hauschild F (1960) *Pharmakologie und Grundlagen der Toxikologie*, 2nd. Thieme, Leipzig, p 568
- Hausmann A, Müller GC (2006) The biblical worms on Jonah's *Ricinus* were *Olepa schleiini* larvae (Lepidoptera, Arctiidae). *Mitt Münch Ent Ges* 95:5–99 ISSN 0340-4943
- He X, McMahon S, Henderson TD II, Griffey SM, Cheng LW (2010) Ricin toxicokinetics and its sensitive detection in mouse sera or feces using immuno-PCR. *PLoS One* 5. <https://doi.org/10.1371/journal.pone.0012858>
- Henry MA, Lesnick E, Westrum LE, Johnson LR, Canfield RC (1987) Ultrastructure of degenerative changes following ricin application to feline dental pulps. *J Neurocytol* 16:601–611
- Herrera L, Bostrom B, Gore L, Sandler E, Lew G, Schlegel PG, Aquino V, Ghetie V, Vitetta ES, Schindler J (2009) A phase 1 study of Combotox in pediatric patients with refractory B-lineage acute lymphoblastic leukemia. *J Pediatr Hematol Oncol* 31:936–941
- Hu R, Zhai Q, Liu W, Liu X (2001) An insight into the mechanism of cytotoxicity of ricin to hepatoma cell: roles of Bcl-2 family proteins, caspases, Ca^{2+} -dependent proteases and protein kinase C. *J Cell Biochem* 81(4):583–593
- Hu WG, Yin J, Chau D, Hu CC, Cherwonogrodzky JW (2014) Anti-ricin protective monoclonal antibodies. *Ricin Toxin*:145–158
- Hughes JN, Lindsay CD, Griffiths GD (1996) Morphology of ricin and abrin exposed endothelial cells is consistent with apoptotic cell death. *Hum Exp Toxicol* 15:443–451
- Inns RH, Tuckwell NJ, Bright JE, Marrs TC (1990) Histochemical demonstration of calcium accumulation in muscle fibres after experimental organophosphate poisoning. *Hum Exp Toxicol* 9: 245–250. <https://doi.org/10.1177/096032719000900407>
- Ishiguro M, Funatsu G, Funatsu M (1971a) Biochemical studies on ricin III. Reinvestigation on the purification of liotin. *Agr Biol Chem* 35: 724–728
- Ishiguro M, Funatsu G, Funatsu M (1971b) Biochemical studies on ricin IV. Amino acid analysis, carboxyl and amino terminal amino acids of ricin D. *Agr Biol Chem* 35:729–733
- Ishiguro M, Mitarai M, Harada H, Sekine I, Nishimori I, Kikutani M (1983) Biochemical studies on oral toxicity of ricin. I Ricin administered orally can impair sugar absorption by rat small intestine. *Chem Pharm Bull (Tokyo)* 31:3222–3227
- Iversen TG, Frerker N, Sandvig K (2012) Uptake of ricinB-quantum dot nanoparticles by a macropinocytosis-like mechanism. *J Nanobiotechnol* 10:33
- Jang SH, Wientjes MG, Lu D, Au JL (2003) Drug delivery and transport to solid tumors. *Pharm Res* 20:1337–1350
- Johnson RC, Lemire SW, Woolfitt SW, Ospina M, Preston KP, Olson CT et al (2005) Quantification of ricinine in rat and human urine: a biomarker for ricin exposure. *J Anal Toxicol* 29:149–155
- Kalb SR, Barr JR (2009) Mass spectrometric detection of ricin and its activity in food and clinical samples. *Anal Chem* 81:2037–2042
- Kalb SR, Schieltz DM, Becher F, Astot C, Fredriksson SA, Barr JR (2015) Recommended mass spectrometry-based strategies to identify ricin-containing samples. *Toxins* 7:4881–4894. <https://doi.org/10.3390/toxins7124854>
- Kobert R (1906) *Lehrbuch der Intoxikationen*, vol 2. Ferdinand Enke, Stuttgart
- Komatsu N, Oda T, Muramatsu T (1998) Involvement of both caspase-like proteases and serine proteases in apoptotic cell death induced by ricin, modeccin, diphtheria toxin, and pseudomonas toxin. *J Biochem* 124:1038–1044
- Korcheva V, Wong J, Corless C, Iordanov M, Magun B (2005) Administration of ricin induces a severe inflammatory response via nonredundant stimulation of ERK, JNK, and P38 MAPK and provides a mouse model of hemolytic uremic syndrome. *Am J Pathol* 166(1):323–339. [https://doi.org/10.1016/S0002-9440\(10\)62256-0](https://doi.org/10.1016/S0002-9440(10)62256-0)
- Korcheva V, Wong J, Lindauer M, Jacoby DB, Iordanov MS, Magun B (2007) Role of apoptotic signaling pathways in regulation of inflammatory responses to ricin in primary murine macrophages. *Mol Immunol* 44(10):2761–2771. <https://doi.org/10.1016/j.molimm.2006.10.025>
- Kumar O, Sugendran K, Vijayaraghavan R (2003) Oxidative stress associated hepatic and renal toxicity induced by ricin in mice. *Toxicol: Off J Int Soc Toxinol* 41:333–338
- Kumar O, Sugendran K, Pant SC, Vijayaraghavan R (2004) Effect of ricin on some biochemical, haematological and histopathological variables. *Def Sci J* 54:493–502
- Lambert JM, Goldmacher VS, Collinson AR, Nadler LM, Blattler WA (1991) An immunotoxin prepared with blocked ricin: a natural plant toxin adapted for therapeutic use. *Cancer Res* 51:6236–6242
- Lappi DA, Kapmeyer W, Beglau JM, Kaplan NO (1978) The disulfide bond connecting the chains of ricin. *Proc Natl Acad Sci U S A* 75: 1096–1100
- Laske DW, Muraszko KM, Oldfield EH, DeVroom HL, Sung C, Dedrick RL, Simon TR, Colandrea J, Copeland C, Katz D, Greenfield L, Groves ES, Houston LL, Youle RJ (1997) Intraventricular immunotoxin therapy for leptomeningeal neoplasia. *Neurosurgery* 41:1039–1049 discussion 1049-1051
- LeMaistre CF, Rosen S, Frankel A, Kornfeld S, Saria E, Meneghetti C, Drajek J, Fishwild D, Scannon P, Byers V (1991) Phase I trial of H65-RTA immunoconjugate in patients with cutaneous T-cell lymphoma. *Blood* 78:1173–1182

- Leshin J, Danielsen M, Credle JJ, Weeks A, O'Connell KP, Dretchen K (2010) Characterization of ricin toxin family members from *Ricinus communis*. *Toxicon* 55:658–661
- Li C, Yan R, Yang Z, Wang H, Zhang R, Chen H, Wang J (2017) BCMab1-Ra, a novel immunotoxin that BCMab1 antibody coupled to ricin A chain, can eliminate bladder tumor. *Oncotarget* 8:46704–46705
- Li Y, Liu W, Sun C, Zheng M, Zhang J, Liu B, Wang Y, Xie Z, Xu N (2018) Hybrids of carbon dots with subunit B of ricin toxin for enhanced immunomodulatory activity. *J Colloid Interface Sci* 523: 226–233
- Lin JY, Liu SY (1986) Studies on the antitumor lectins isolated from the seeds of *Ricinus communis* (castor bean). *Toxicon* 24(8):757–765
- Lin JY, Tserng KY, Chen CC, Lin LT, Tung TC (1970) Abrin and ricin: new anti-tumour substances. *Nature* 227(18):292–293
- Lin JY, Chang YC, Huang LY, Tung TC (1973) The cytotoxic effects of abrin and ricin on Ehrlich ascites tumor cells. *Toxicon: Off J Int Soc Toxinol* 11:379–381
- Lindauer M, Wong J, Magun B (2010) Ricin toxin activates the NALP3 inflammasome. *Toxins (Basel)* 2(6):1500–1514. <https://doi.org/10.3390/toxins2061500>
- Ling EA, Wen CY, Shieh JY, Yich TY, Wong WC (1991) Ultrastructural changes of the nodose ganglion cells following an intraneural injection of *Ricinus communis* agglutinin-60 into the vagus nerve in hamsters. *J Anat* 179:23–32
- Lopez Nunez OF, Pizon AF, Tamama K (2017) Ricin poisoning after oral ingestion of castor beans: a case report and review of the literature and laboratory testing. *J Emerg Med* 53(5):e67–e71. <https://doi.org/10.1016/j.jemermed.2017.08.023>
- Lopez MAR, Perez GS, Hernandez CR, Fefer PG, Sanchez MAZ (2010) Activity of *Ricinus communis* (Euphorbiaceae) against *Spodoptera frugiperda* (Lepidoptera: Noctuidae). *Afr J Biotechnol* 9:1359–1365
- Lord JM, Spooner RA (2011) Ricin trafficking in plant and mammalian cells. *Toxins* 3:787–801
- Lord JM, Roberts LM, Robertus D (1994) Ricin: structure, mode of action, and some current applications. *FASEB J* 8:201–208
- LoRusso PM, Lomen PL, Redman BG, Poplin E, Bander JJ, Valdivieso M (1995) Phase I study of monoclonal antibody-ricin A chain immunoconjugate Xomazyme-791 in patients with metastatic colon cancer. *Am J Clin Oncol* 18:07–312
- Lynch TJ Jr, Lambert JM, Coral F, Shefner J, Wen P, Blattler WA, Collinson AR, Ariniello PD, Braman G, Cook S, Esseltine D, Elias A, Skarin A, Ritz J (1997) Immunotoxin therapy of small-cell lung cancer: a phase I study of N901-blocked ricin. *J Clin Oncol* 15:723–734
- Ma L, Hsu CH, Patterson E, Thadani U, Robinson CP (1996) Ricin depresses cardiac function in the rabbit heart. *Toxicol Appl Pharmacol* 138(1):72–76. <https://doi.org/10.1006/taap.1996.0099>
- Madan S, Ghosh PC (1992) Enhancing potency of liposomal monensin on ricin cytotoxicity in mouse macrophage tumor cells. *Biochem Int* 28:287–295
- Madan S, Vasandani VM, Ghosh PC (1993) Potentiation of ricin cytotoxicity by liposomal monensin under *in vitro* and *in vivo* conditions. *Indian J Biochem Biophys* 30:405–410
- Magnusson S, Kjekshus R, Berg T (1993) Characterization of two distinct pathways of endocytosis of ricin by rat liver endothelial cells. *Exp Cell Res* 205(1):118–125. <https://doi.org/10.1006/excr.1993.1065>
- Manjunatha DH, Chua LS (2014) The anti-inflammatory and wound healing properties of honey. *Eur Food Res Technol* 239(6):1003–1014. <https://doi.org/10.1007/s00217-014-2297-6>
- Mensah MB, Awudza JAM, O'Brien P (2018) Castor oil: a suitable green source of capping agent for nanoparticle syntheses and facile surface functionalization. *R Soc Open Sci* 5:180824. <https://doi.org/10.1098/rsos.180824>
- Messmann RA, Vitetta ES, Headlee D, Senderowicz AM, Figg WD, Schindler J, Michiel DF, Creekmore S, Steinberg SM, Kohler D, Jaffe ES, Stetler-Stevenson M, Chen H, Ghetie V, Sausville EA (2000) A phase I study of combination therapy with immunotoxins IgG-HD37-deglycosylated ricin A chain (dgA) and IgG-RFB4-dgA (Combotox) in patients with refractory CD19(+), CD22(+) B cell lymphoma. *Clin Cancer Res* 6:1302–1313
- Mise T, Funatsu G, Ishiguro M, Funatsu M (1977) Isolation and characterization of ricin E from castor beans. *Agric Biol Chem* 41(10): 2041–2046. <https://doi.org/10.1271/bbb1961.41.2041>
- Morlon-Guyot J, Helmy M, Lombard-Frasca S, Pignol D, Pieroni G, Beaumelle B (2003) Identification of the ricin lipase site and implication in cytotoxicity. *J Biol Chem* 278:17006–17011
- Mosinger M (1951) Necrosing or clastic effects of ricin on different organs and on experimental sarcomas. *C R Seances Soc Biol Fil* 145:412–415
- Mutlu H, Meier MAR (2010) Castor oil as a renewable resource for the chemical industry. *Eur J Lipid Sci Technol* 112:10–30
- Muto S, Beevers H (1974) Lipase activities in castor bean endosperm during germination. *Plant Physiol* 54(1):23–28
- Nascimento VV, Castro HC, Abreu PA, Oliveira AEA, Fernandez JH, Araujo JS, Machado OLT (2011) In silico structural characteristics and α -amylase inhibitory properties of Ric c 1 and Ric c 3, allergenic 2s albumins from *Ricinus communis* seeds. *J Agric Food Chem* 59:4814–4821. <https://doi.org/10.1021/jf104638b>
- Neville DM Jr, Youle RJ (1982) Monoclonal antibody-ricin or ricin A chain hybrids: kinetic analysis of cell killing for tumor therapy. *Immunol Rev* 62:75–91
- Nicolson GL, Poste G (1978) Mechanism of resistance to ricin toxin in selected mouse lymphoma cell lines. *J Supramol Struct* 8:235–245
- Noy-Porat T, Alcalay R, Epstein E, Sabo T, Kronman C, Mazor O (2017) Extended therapeutic window for post-exposure treatment of ricin intoxication conferred by the use of high-affinity antibodies. *Toxicon* 127:100–105. <https://doi.org/10.1016/j.toxicon.2017.01.009> PMID: 28089771
- Ogunniyi DS (2006) Castor oil: a vital industrial raw material. *Bioresour Technol* 97(9):1086–1091
- Olaifa JI, Matsumura F, Zeevaart JAD, Mullin CA, Charalambous P (1991) Lethal amounts of ricinine in green peach aphids (*Myzus persicae*) (Suzler) fed on castor bean plants. *Plant Sci* 73(2):253–256
- Olsnes S (2004) The history of ricin, abrin and related toxins. *Toxicon: Off J Int Soc Toxinol* 44:361–370
- Olsnes S, Pihl A (1973a) Different biological properties of the two constituent peptide chains of ricin, a toxic protein inhibiting protein synthesis. *Biochemistry* 12(16):3121–3126
- Olsnes S, Pihl A (1973b) Isolation and properties of abrin: a toxic protein inhibiting protein synthesis evidence for different biological functions of its two constituent-peptide chains. *Eur J Biochem* 35:179–185 PMID:4123356
- Olsnes S, Saltvedt E, Pihl A (1974) Isolation and comparison of galactose-binding lectins from *Abrus precatorius* and *Ricinus communis*. *J Biol Chem* 249:803–810
- Ory RL, Yatsu LY, Kircher HW (1968) Association of lipase activity with the spherulines of *Ricinus communis*. *Arch Biochem* 123:255–264. [https://doi.org/10.1016/0003-9861\(68\)90132-X](https://doi.org/10.1016/0003-9861(68)90132-X)
- Pabis S, Kula J (2016) Synthesis and bioactivity of (R)-ricinoleic acid derivatives: a review. *Curr Med Chem* 23(35):4037–4056
- Patel VR, Dumancas GG, Kasi Viswanath LC, Maples R, Subong BJJ (2016) Castor oil: properties, uses, and optimization of processing parameters in commercial production. *Lipid Insights* 9:1–12. <https://doi.org/10.4137/LPI.S40233>
- Patocka J (2018) Highly toxic ribosome-inactivating proteins as chemical warfare or terrorist agents. *Mil Med Sci Lett (Voj Zdrav Listy)* 87: 1–11 ISSN 0372-7025

- Pecot CV, Calin GA, Coleman RL, Lopez-Berestein G, Sood AK (2011) RNA interference in the clinic: challenges and future directions. *Nat Rev Cancer* 11:59–67
- Pittman CT, Guido J, Hamelin EI, Blake TA, Johnson RC (2013) Analysis of a ricin biomarker, ricinine, in 989 individual human urine samples. *J Anal Toxicol* 37(4):237–240. <https://doi.org/10.1093/jat/bkt010>
- Poelchen W, Wirkner K (2003) Ein potenzieller biologischer Kampfstoff. *Pharmazeutische Zeitung online* (06/2003)
- Preijers FW, Tax WJ, De Witte T, Janssen A, vd Heijden H, Vidal H, Wessels JM, Capel PJ (1988) Relationship between internalization and cytotoxicity of ricin A-chain immunotoxins. *Br J Haematol* 70: 289–294
- Press OW, Vitetta ES, Farr AG, Hansen JA, Martin PJ (1986) Evaluation of ricin A-chain immunotoxins directed against human T cells. *Cell Immunol* 102:10–20
- Prigent J, Panigai L, Lamourette P, Sauvaire D, Devilliers K et al (2011) Neutralising antibodies against ricin toxin. *PLoS One* 6(5):e20166. <https://doi.org/10.1371/journal.pone.0020166>
- Ramsden CS, Drayson MT, Bell EB (1989) The toxicity, distribution and excretion of ricin holotoxin in rats. *Toxicology* 55(1–2):161–171
- Rana M, Dhamija H, Prashar B, Sharma S (2012) *Ricinus communis* L. – a review. *Int J Pharmacol Res* 4(4):1706–1711
- Rao PV, Jayaraj R, Bhaskar AS, Kumar O, Bhattacharya R, Saxena P, Dash PK, Vijayaraghavan R (2005) Mechanism of ricin-induced apoptosis in human cervical cancer cells. *Biochem Pharmacol* 69: 855–865
- Rathore SS, Ghosh PC (2008) Effect of surface charge and density of distearylphosphatidylethanolamine-mPEG-2000 (DSPE-mPEG-2000) on the cytotoxicity of liposome-entrapped ricin: effect of lysosomotropic agents. *Int J Pharm* 350:79–94
- Tchou-Wong KM, Roy CJ, Parent C, Cabrera M, Guillemin J, Mac Loughlin R, Levacher E, Fontayne A, Douziech-Eyrolles L, Junqua-Moulet A, Guilleminault L, Thullier P, Guillot-Combe E, Vecellio L, Heuzé-Vourc'h N (2016) Development of a drug delivery system for efficient alveolar delivery of a neutralizing monoclonal antibody to treat pulmonary intoxication to ricin. *J Control Release* 234:21–32. <https://doi.org/10.1016/j.jconrel.2016.05.018>
- Ribeiro BD, Machado de Castro A, Zarur Coelho MA, Guimarães Freire DM (2011) Production and use of lipases in bioenergy: a review from the feedstocks to biodiesel production. *Enzym Res*:615803. <https://doi.org/10.4061/2011/615803>
- Robert Koch-Institut (2017) RKI-Ratgeber für Ärzte: Rizin-Intoxikation. *Epid Bull* 32:315–321. <https://doi.org/10.17886/EpiBull-2017-042>
- Roberts LM, Lamb FI, Pappin DJ, Lord JM (1985) The primary sequence of *Ricinus communis* agglutinin. Comparison with ricin. *J Biol Chem* 260:15682–15686
- RÖMPP Lexikon Chemie, 10. Auflage (1996–1999) Band 2: Cm – G. Herausgegeben von Falbe J, Regitz M. Georg Thieme Verlag
- Rosenfeld R, Alcalay R, Mechaly A, Lapidot G, Epstein E, Kronman C, Fleishman SJ, Mazor O (2017) Improved antibody-based ricin neutralization by affinity maturation is correlated with slower off-rate values. *Protein Eng Des Sel* 30(9):611–617. <https://doi.org/10.1093/protein/gzx028> PMID: 28472478
- Roth L, Daunderer M, Kormann K (2008) Giftpflanzen – Pflanzengifte. Vorkommen, Wirkung, Therapie. Allergische und phototoxische Reaktionen, 5. Aufl. Hamburg
- Roxas-Duncan VI, Smith LA (2012) Ricin perspective in bioterrorism, Dr. Stephen Morse (Ed.) ISBN:978-953-51-0205-2
- Roy CJ, Hale M, Hartings JM, Pitt L, Duniho S (2003) Impact of inhalation exposure modality and particle size on the respiratory deposition of ricin in BALB/c mice. *Inhal Toxicol* 15:619–638. <https://doi.org/10.1080/08958370390205092>
- Roy CJ, Song K, Sivasubramani SK, Gardner DJ, Pincus SH (2012) Animal models of ricin toxicosis. *Curr Top Microbiol Immunol* 357:243–257. https://doi.org/10.1007/82_2011_173
- Sandvig K, Skotland T, van Deurs B, Klokke TI (2013) Retrograde transport of protein toxins through the Golgi apparatus. *Histochem Cell Biol* 140:317–326. <https://doi.org/10.1007/s00418-013-1111-z>
- Sausville EA, Headlee D, Stetler-Stevenson M, Jaffe ES, Solomon D, Figg WD, Herdt J, Kopp WC, Rager H, Steinberg SM et al (1995) Continuous infusion of the anti-CD22 immunotoxin IgG-RFB4-SMPT-dgA in patients with B-cell lymphoma: a phase I study. *Blood* 85:3457–3465
- Scarpa A, Guerci A (1982) Various uses of the castor oil plant (*Ricinus communis* L.): a review. *J Ethnopharmacol* 5:117–137
- Schindler J, Gajavelli S, Ravandi F, Shen Y, Parekh S, Braunschweig I, Barta S, Ghetie V, Vitetta E, Verma A (2011) A phase I study of a combination of anti-CD19 and anti-CD22 immunotoxins (Combotox) in adult patients with refractory B-lineage acute lymphoblastic leukaemia. *Br J Haematol* 154:471–476
- Schnell R, Staak O, Borchmann P, Schwartz C, Matthey B, Hansen H, Schindler J, Ghetie V, Vitetta ES, Diehl V, Engert A (2002) A phase I study with an anti-CD30 ricin A-chain immunotoxin (Ki-4.dgA) in patients with refractory CD30+ Hodgkin's and non-Hodgkin's lymphoma. *Clin Cancer Res* 8:1779–1786
- Schnell R, Borchmann P, Staak JO, Schindler J, Ghetie V, Vitetta ES, Engert A (2003) Clinical evaluation of ricin A-chain immunotoxins in patients with Hodgkin's lymphoma. *Ann Oncol* 14:729–736
- Scholl R (2002) Der Papyrus Ebers. Die größte Buchrolle zur Heilkunde Ägyptens, Leipzig (Schriften aus der Universitätsbibliothek Leipzig, 7).
- Sehgal P, Khan M, Kumar O, Vijayaraghavan R (2010) Purification, characterization and toxicity profile of ricin isoforms from castor beans. *Food Chem Toxicol* 48:3171–3176. <https://doi.org/10.1016/j.fct.2010.08.015>
- Sehnke PC, Ferl RJ (1999) Processing of preproricin in transgenic tobacco. *Protein Expr Purif* 15:188–195. <https://doi.org/10.1006/prep.1998.0993>
- Sehnke PC, Pedrosa L, Paul AL, Frankel AE, Ferl RJ (1994) Expression of active, processed ricin in transgenic tobacco. *J Biol Chem* 269(36):22473–22476
- Selvaggi K, Saria EA, Schwartz R, Vlock DR, Ackerman S, Wedel N, Kirkwood JM, Jones H, Ernstoff MS (1993) Phase I/II study of murine monoclonal antibody-ricin A chain (XOMAZYME-Mel) immunoconjugate plus cyclosporine A in patients with metastatic melanoma. *J Immunother Emphasis Tumor Immunol: Off J Soc Biol Ther* 13:201–207
- Severino LS, Auld DL, Baldanzi M et al (2012) A review on the challenges for increased production of castor. *Agron J* 104(4):853. <https://doi.org/10.2134/agronj2011.0210> https://www.researchgate.net/publication/273669791_A_Review_on_the_Challenges_for_Increased_Production_of_Castor. Accessed 18 March 2019
- Sha O, Yew DT, Ng TB, Yuan L, Kwong WH (2010) Different in vitro toxicities of structurally similar type I ribosome-inactivating proteins (RIPs). *Toxicol in Vitro* 24(4):1176–1182. <https://doi.org/10.1016/j.tiv.2010.02.014>
- Shaik MS, Ikediobi O, Tumage VD, McSweeney J, Kanikkannan N, Singh M (2001) Long-circulating monensin nanoparticles for the potentiation of immunotoxin and anticancer drugs. *J Pharm Pharmacol* 53: 617–627
- Simmons BM, Stahl PD, Russell JH (1986) Mannose receptor-mediated uptake of ricin toxin and ricin A chain by macrophages. Multiple intracellular pathways for a chain translocation. *J Biol Chem* 261(17):7912–7920. PMID 3711116
- Singh A, Kaur J (2017) Activity of foliage extracts of *Ricinus communis* L. (Euphorbiaceae) against myiasis causing larvae of *Chrysomya*

- bezziana Villeneuveae (Diptera: Calliphoridae). J Vet Med Res 4(1): 1070
- Singh M, Griffin T, Salimi A, Micetich RG, Atwal H (1994) Potentiation of ricin A immunotoxin by monoclonal antibody targeted monensin containing small unilamellar vesicles. Cancer Lett 84:15–21
- Singh M, Ferdous AJ, Kanikkannan N, Faulkner G (2001) Stealth monensin immunoliposomes as potentiator of immunotoxins *in vitro*. Eur J Pharm Biopharm 52:13–20
- Slominska-Wojewodzka M, Sandvig K (2013) Ricin and ricin-containing immunotoxins: insights into intracellular transport and mechanism of action *in vitro*. Antibodies 2:236–269
- Smallshaw JE, Richardson JA, Pincus S, Schindler J, Vitetta ES (2005) Preclinical toxicity and efficacy testing of RiVax, a recombinant protein vaccine against ricin. Vaccine 23:4775–4784. <https://doi.org/10.1016/j.vaccine.2005.04.037>
- Smith DC, Salio M, Lord JM, Roberts LM, Cerundolo V (2004) Lack of dendritic cell maturation by the plant toxin ricin. Eur J Immunol 34(8):2149–2157. <https://doi.org/10.1002/eji.200425157>
- Soler-Rodríguez AM, Ghetie MA, Oppenheimer-Marks N, Uhr JW, Vitetta ES (1993) Ricin A-chain and ricin A-chain immunotoxins rapidly damage human endothelial cells: implications for vascular leak syndrome. Exp Cell Res 206(2):227–234
- Sousa NL, Cabral GB, Vieira PM, Balsoni AB, Aragao FJL (2017) Sci Rep 7:15385. <https://doi.org/10.1038/s41598-017-15636-7>
- Sparapani M, Buonamici L, Ciani E, Battelli MG, Ceccarelli G, Stirpe F, Contestabile A (1997) Toxicity of ricin and volkensin, two ribosome-inactivating proteins, to microglia, astrocyte, and neuron cultures. Glia 20(3):203–209
- Spies JR, Coulson EJ (1964) The chemistry of allergens: XVI. Ion exchange fractionation of the castor bean allergen, CB-1A, and antigenic specificity relationships of the fractions. J Biol Chem 239: 1818–1827
- Spitler LE, del Rio M, Khentigan A, Wedel NI, Brophy NA, Miller LL, Harkonen WS, Rosendorf LL, Lee HM, Mischak RP et al (1987) Therapy of patients with malignant melanoma using a monoclonal antimelanoma antibody-ricin A chain immunotoxin. Cancer Res 47: 1717–1723
- Srivastava A, Mathur RM, Prakash R, Agrawal S (2016) Studies on Ricinus lipase enzyme isolated from castor seeds. Orient J Chem 32(2):1235–1247. <https://doi.org/10.13005/ojc/320250>
- Stern D, Skiba M, Kampa B, Worbs S, Dorner BG (2018) Anforderungen an Rizin-Nachweismethoden zur Detektion und Identifizierung aus Verdachtsproben. Toxichem Krimtech 85(3):100
- Stillmark H (1888) Ueber Ricin, ein giftiges Fragment aus den Samen von Ricinus comm. L und einigen anderen Euphorbiaceen; PhD Thesis, Kaiserliche Universität zu Dorpat: Tartu, Estonia
- Stone MJ, Sausville EA, Fay JW, Headlee D, Collins RH, Figg WD, Stetler-Stevenson M, Jain V, Jaffe ES, Solomon D, Lush RM, Senderowicz A, Ghetie V, Schindler J, Uhr JW, Vitetta ES (1996) A phase I study of bolus versus continuous infusion of the anti-CD19 immunotoxin, IgG-HD37-dgA, in patients with B-cell lymphoma. Blood 88:1188–1197
- Streit WJ, Kreutzberg GW (1988) Response of endogenous glial cells to motor neuron degeneration induced by toxic ricin. J Comp Neurol 268(2):248–263. <https://doi.org/10.1002/cne.902680209>
- Tarsio JF, Kelleher PJ, Tarsio M, Emery JM, Lam DM (1997) Inhibition of cell proliferation on lens capsules by 4197X-ricin A immunoconjugate. J Cataract Refract Surg 23(2):260–266
- Taubenschmid J, Stadlmann J, Jost M, Klock TI, Rillahan CD, Leibbrandt A, Mechtler K, Paulson JC, Jude J, Zuber J, Sandvig K, Elling U, Marquardt T, Thiel C, Koerner C, Penninger JM (2017) A vital sugar code for ricin toxicity. Cell Res 27(11):1351–1364. <https://doi.org/10.1038/cr.2017.116>
- Taylor S, Massiah A, Lomonosoff G, Roberts LM, Lord JM, Hartley M (1994) Correlation between the activities of five ribosome-inactivating proteins in depurination of tobacco ribosomes and inhibition of tobacco mosaic virus infection. Plant J 5:827–835
- Teuscher E, Lindequist U (2010) Biogene Gifte (Biologie-Chemie-Pharmakologie-Toxikologie), Wissenschaftliche Verlagsgesellschaft mbH Stuttgart; 3. Auflage. ISBN 978-3-8047-2438-9
- Thiermann H, Zöller L, Szinicz (2013) Chemische und biologische Kampfstoffe. In: Marquardt H, Schäfer SG, Barth H (eds) Toxikologie. Wissenschaftliche Verlagsgesellschaft Stuttgart, 3. Auflage, pp 915–954 ISBN 978-3-8047-2876-9
- Thorpe SC, Kemeny DM, Panzani RC, McGurl B, Lord M (1988) Allergy to castor bean II Identification of the major allergens in castor bean seeds. J Allergy Clin Immunol 82:67–72
- Timar J, McIntosh DP, Henry R, Cumber AJ, Pamell GD, Davies AJ (1991) The effect of ricin B chain on the intracellular trafficking of an A chain immunotoxin. Br J Cancer 64:655–662
- Tokuyama T, Daly J, Witkop B (1969) The structure of batrachotoxin, a steroidal alkaloid from the Colombian arrow poison frog, Phyllobates aurotaenia, and partial synthesis of batrachotoxin and its analogs and homologs. J Am Chem Soc 91(14):3931–3938. <https://doi.org/10.1021/ja01009a052>
- Tong WM, Sha O, Ng TB, Cho EY, Kwong WH (2012) Different in vitro toxicity of ribosome-inactivating proteins (RIPs) on sensory neurons and Schwann cells. Neurosci Lett 524(2):89–94. <https://doi.org/10.1016/j.neulet.2012.07.006>
- Törnquist E, Liu L, Mattsson P, Svensson M (1997) Response of glial cells and activation of complement following motoneuron degeneration induced by toxic ricin. Neurosci Res 28:167–175
- Toth RT, Angalakurthi SK, Van Slyke G, Vance DJ, Hickey JM, Joshi SB, Middaugh CR, Volkin DB, Weis DD, Mantis NJ (2017) High-definition mapping of four spatially distinct neutralizing epitope clusters on RiVax, a candidate ricin toxin subunit vaccine. Clin Vaccine Immunol 24(12):e00237–17. <https://doi.org/10.1128/CVI.00237-17>
- Tunaru S, Althoff TF, Nüsing RM, Diener M, Offermanns S (2012) Castor oil induces laxation and uterus contraction via ricinoleic acid activating prostaglandin E 3 receptors. PNAS. <https://doi.org/10.1073/pnas.1201627109>
- Tyagi N, Ghosh PC (2011) Folate receptor mediated targeted delivery of ricin entrapped into sterically stabilized liposomes to human epidermoid carcinoma (KB) cells: effect of monensin intercalated into folate-tagged liposomes. Eur J Pharm Sci 43:343–353
- Tyagi N, Rathore SS, Ghosh PC (2011) Enhanced killing of human epidermoid carcinoma (KB) cells by treatment with ricin encapsulated into sterically stabilized liposomes in combination with monensin. Drug Deliv 18:394–404
- Tyagi N, Rathore SS, Ghosh PC (2013) Efficacy of liposomal monensin on the enhancement of the antitumour activity of liposomal ricin in human epidermoid carcinoma (KB) cells. Indian J Pharm Sci 75:16–22
- Tyagi N, Tyagi M, Pachauri M, Ghosh PC (2015) Potential therapeutic applications of plant toxin-ricin in cancer: challenges and advances. Tumour Biol 36:8239–8246
- Upasani SM, Kotkar HM, Mendki PS, Maheshwari VL (2003) Partial characterization and insecticidal properties of *Ricinus communis* L. foliage flavonoids. Pest Manag Sci 59:1349–1354
- van Horssen PJ, van Oosterhout YV, de Witte T, Preijers FW (1995) Cytotoxic potency of CD22-ricin A depends on intracellular routing rather than on the number of internalized molecules. Scand J Immunol 41:563–569
- Vandenborre G, Smagghe G, Van Damme EJM (2011) Plant lectins as defense proteins against phytophagous insects. Phytochemistry 72: 1538–1550. <https://doi.org/10.1016/j.phytochem.2011.02.024>

- Vasandani VM, Madan S, Ghosh PC (1992) *In vivo* potentiation of ricin toxicity by monensin delivered through liposomes. *Biochim Biophys Acta* 1116:315–323
- Verougstraete N, Helsloot D, Deprez C, Heylen O, Casier I, Croes K (2018) Lethal injection of a castor bean extract: ricinine quantification as a marker for ricin exposure using a validated LC-MS/MS method. *J Anal Toxicol*. <https://doi.org/10.1093/jat/bky100>
- Vieira C, Evangelista S, Cirillo R, Lippi A, Maggi CA, Manzini S (2000) Effect of ricinoleic acid in acute and subchronic experimental models of inflammation. *Mediators Inflamm* 9(5):223–228. <https://doi.org/10.1080/09629350020025737>
- Vijayan K, Anuradha HJ, Nair CV, Pradeep AR, Awasthi AK, Saratchandra B, Rahman SAS, Singh KC, Chakraborti R, Urs SR (2006) Genetic diversity and differentiation among populations of the Indian eri silkworm, *Samia cynthia ricini*, revealed by ISSR markers. *J Insect Sci* 6:30 (online: [insectscience.org/6.30])
- Vitetta E (2006) Biomedical and biodefense uses for ricin. An [ActionBioscience.org](http://www.actionbioscience.org) original interview. Accessed 01.02.2019, available: from <http://www.actionbioscience.org/biotechnology/vitetta.html>
- Vitetta ES, Stone M, Amlot P, Fay J, May R, Till M, Newman J, Clark P, Collins R, Cunningham D et al (1991) Phase I immunotoxin trial in patients with B-cell lymphoma. *Cancer Res* 51:4052–4058
- Vitetta ES, Smallshaw JE, Coleman E et al (2005) A pilot clinical trial of a recombinant ricin vaccine in normal humans. *PNAS* 103:2268–2273 ISSN (printed): 0027-8424 ISSN (electronic)
- Vitetta ES, Smallshaw JE, Coleman E, Jafri H, Foster C, Munford R, Schindler J (2006) A pilot clinical trial of a recombinant ricin vaccine in normal humans. *Proc Natl Acad Sci U S A* 103:2268–2273
- von Fischer-Benzon R (1894) *Altdeutsche Gartenflora. Untersuchungen über die Nutzpflanzen des deutschen Mittelalters, ihre Wanderung und ihre Vorgeschichte im klassischen Altertum*. Kiel und Leipzig, Verlag von Lipsius & Tischer <https://www.biodiversitylibrary.org/item/48587#page/9/mode/1up>. Accessed 18 March 2019
- Vyleta ML, Wong J, Magun BE (2012) Suppression of ribosomal function triggers innate immune signaling through activation of the NLRP3 inflammasome. *PLoS One* 7(5):e36044. <https://doi.org/10.1371/journal.pone.0036044>
- Wachira SW, Omar S, Jacob JW, Wahome M, Alborn HT, Spring DR et al (2014) Toxicity of six plant extracts and two pyridone alkaloids from *Ricinus communis* against the malaria vector *Anopheles gambiae*. *Parasit Vectors* 7(1):312–319
- Waller GR, Skursky L (1972) Translocation and metabolism of ricinine in the castor bean plant, *Ricinus communis* L. *Plant Physiol* 50(5):622–626
- Watanabe Y, Osawa T (1987) Effect of ricin or ricin A-chain encapsulated in anti-carcinoembryonic antigen (CEA) antibody-bearing liposomes on CEA-producing tumor cells. *Chem Pharm Bull* 35:740–747
- Wawrzyniczak EJ, Watson GJ, Cumber AJ, Henry RV, Parnell GD, Rieber EP, Thorpe PE (1991) Blocked and non-blocked ricin immunotoxins against the CD4 antigen exhibit higher cytotoxic potency than a ricin A chain immunotoxin potentiated with ricin B chain or with a ricin B chain immunotoxin. *Cancer Immunol Immunother* 32(5):289–295
- Weber M (2014) The use of ricin abrin or modeccin in the context of criminal or terrorist attacks. <http://www.cbmeportal.com/the-use-of-ricin-abrin-or-modeccin-in-the-context-of-criminal-or-terrorist-attacks>. Accessed 18 March 2019
- Weber M (2018) Bedrohungspotential des Bio-Kampfstoffes Rizin. *Toxichem Krimtech* 85(3):97–99
- Weber M, Schulz H (2011) Immunological detection of ricin and castor seeds in beverages. *Food Consum Prod Toxichem Krimtech* 78(Special Issue):276
- Weller U, Dauzenroth ME, Meyer zu Heringdorf D, Habermann E (1989) Chains and fragments of tetanus toxin. Separation, reassociation and pharmacological properties. *Eur J Biochem* 182:649–656
- Weng X, Wang M, Ge J, Yu S, Liu B, Zhong J, Kong J (2009) Carbon nanotubes as a protein toxin transporter for selective HER2-positive breast cancer cell destruction. *Mol Biosyst* 5: 1224–1231
- Westendorf J (2013) *Naturstoffe*. In: Marquardt H, Schäfer SG, Barth H, Toxikologie. Wissenschaftliche Verlagsgesellschaft Stuttgart, 3. Auflage, 1133. ISBN 978-3-8047-2876-9
- Worbs S, Köhler K, Pauly D, Avondet MC, Schaer M, Dörner MB, Dörner BG (2011) *Ricinus communis* intoxications in human and veterinary medicine—a summary of real cases. *Toxins* 3(10):1332–1372. <https://doi.org/10.3390/toxins3101332>
- Worbs S, Skiba M, Söderström M, Rapinaja ML, Zeleny R, Russmann H, Schimmel H, Vanninen P, Fredriksson SA, Dörner BG (2015) Characterization of ricin and *R. communis* agglutinin reference materials. *Toxins* 7:4906–4934. <https://doi.org/10.3390/toxins7124856>
- Wu YH, Shih SF, Lin JY (2004) Ricin triggers apoptotic morphological changes through caspase-3 cleavage of BAT3. *J Biol Chem* 279: 19264–19275
- Wu F, Fan S, Martiniuk F, Pincus S, Muller S, Kohler H, Tchou-Wong KM (2010) Protective effects of anti-ricin A-chain antibodies delivered intracellularly against ricin-induced cytotoxicity. *World J Biol Chem* 1:188–195
- Xu N, Yuan H, Liu W, Li S, Liu Y, Wan J, Li X, Zhang R, Chang Y (2013) Activation of RAW264.7 mouse macrophage cells in vitro through treatment with recombinant ricin toxin-binding subunit B: involvement of protein tyrosine, NF- κ B and JAK-STAT kinase signaling pathways. *Int J Mol Med* 32(3):729–735. <https://doi.org/10.3892/ijmm.2013.1426>
- Zahir AA, Rahuman AA, Bagavan A et al (2010) Evaluation of botanical extracts against *Haemaphysalis bispinosa* Neumann and *Hippobosca maculata* Leach. *Parasitol Res* 107(3):585–592. <https://doi.org/10.1007/s00436-010-1898-7>
- Zhan J, Zhou P (2003) A simplified method to evaluate the acute toxicity of ricin and ricinus agglutinin. *Toxicology* 186:119–123
- Zhang L, Hsu CH, Robinson CP (1994) Effects of ricin administration to rabbits on the ability of their coronary arteries to contract and relax in vitro. *Toxicol Appl Pharmacol* 129(1):16–22. <https://doi.org/10.1006/taap.1994.1224>
- Zhang J, Qin DA, Cheng BX, Ru XQ, Li H, Li L, Yang HP (1999) Preparation, characteristics and toxicity of ricin-containing galactosyl ceramide liposomes. *Sheng Wu Hua Xue Yu Sheng Wu Wu Li Xue Bao (Shanghai)*. *Acta biochimica et biophysica Sinica* 1999;31(4):472–474. <https://www.ncbi.nlm.nih.gov/pubmed.12115009>

Final report on the safety assessment of *Ricinus communis* (Castor) seed oil, hydrogenated castor oil, glyceryl ricinoleate, glyceryl ricinoleate se, ricinoleic acid, potassium ricinoleate, sodium ricinoleate, zinc ricinoleate, cetyl ricinoleate, ethyl ricinoleate, glycol ricinoleate, isopropyl ricinoleate, methyl ricinoleate, and octyldodecyl ricinoleate (2007) *Int J Toxicol*. 26 Suppl 3:31–77. <https://doi.org/10.1080/10915810701663150>

Patent (1983) (US 4398937, "Selective algacides for control of cyanochloronta"(1983 Date of Patent)). <https://patents.justia.com/patent/4398937>. Accessed 23 July 2019.

<https://www.naturinstitut.info/rizinusoel.html>; Accessed 18 March 2019

- “Giftpflanze des Jahres 2018”; Botanischer Sondergarten Wandsbek: <https://www.hamburg.de/wandsbek/giftpflanze-des-jahres>; Accessed 18 March 2019
- <https://emergency.cdc.gov/agent/ricin/facts.asp>; Accessed 18 March 2019a
- <https://www.opcw.org/our-work/what-chemical-weapon>; Accessed 29 March 2019b
- Clutal Omega: <https://www.ebi.ac.uk/services>; Accessed 29 March 2019
- <https://www.caymanchem.com/msdss/26384m.pdf> (2019); Accessed 09 May 2019c
- European Medicines Agency (2012): Assessment report on *Viscum album* L., herba (2012); EMA/HMPC/246778/2009; Committee on Herbal Medicinal Products (HMPC); https://www.ema.europa.eu/en/documents/herbal-report/final-assessment-report-viscum-album-l-herba_en.pdf; Accessed 01 July 2019d

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