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PHARMACOLOGY OF *TITYUS SERRULATUS* SCORPION VENOM

Lineu Freire-Maia, Departamento de Farmacologia. ICB-UFMG. Belo Horizonte, MG.

ABSTRACT: The data presented in this lecture seem to indicate that the effects evoked by scorpion toxins, obtained from *Tityus serrulatus* scorpion venom are due to actions on specific sites of sodium channels, with a subsequent depolarization of the membranes. The respiratory arrhythmias are due, at least in part, to depolarizations of visceral afferent fibers, induced by the toxin, whereas the other effects are mainly due to release from nerve endings of chemical mediators, such as acetylcholine and catecholamines.
KEYWORDS: Scorpion venom; pharmacology.

INTRODUCTION

Scientists all over the world have been interested in scorpion venoms because of the high incidence of fatal cases of scorpion poisoning.[21] I will describe in this lecture the pharmacology of the venom obtained from the South American scorpion *Tityus serrulatus* Lutz & Mello 1922.

Cardiovascular effects. Intravenous injection of crude venom or purified toxins[29] obtained from *T. serrulatus* scorpion venom in rats or dogs induced an acute arterial hypertension, due to the release of catecholamines from adrenal glands and postganglionic nerve endings: the catecholamines acting on alpha adrenergic receptors would increase the peripheral resistence; at least, the catecholamines could act also on beta adrenergic receptors, either increasing the cardiac contractility or releasing renin from the kidneys[1,10,35,45].

Scorpion toxin also induces complex cardiac arrhythmias in the rat. Bilateral cervical vagotomy does not prevent the production of sinus bradycardia, SA block or AV block, after toxin injection; in vagotomized rats, physostigmine enhances,

hexamethonium decreases and atropine abolishes the bradycardiac effects. Moreover, propranolol prevents or abolishes the sinus tachycardia and changes the ventricular into a sinus rhythm. Based on the data we have concluded that the sinus bradycardia, SA and AV block are due to release of acetylcholine by actions of the toxin on vagal ganglia and postganglionic nerve endings in the heart whereas the sinus tachycardia, the ventricular ectopic beats and the idioventricular rhythm are caused by activation of beta adrenergic receptors in the heart by catecholamines released by the toxin.[19,23]

The crude venom of the scorpion *Tityus serrulatus* evokes, in isolated guinea pig heart, a short-lasting bradycardia followed by a conspicuous increase in the force and the rate of cardiac contractions, due to a local release of acetylcholine and noradrenaline[10]. A direct demonstration of the release of noradrenaline in isolated guinea pig atria by tityustoxin, extracted from the venom of *Tityus serrulatus*[29] was also reported[36].

Experiments performed by our group in isolated guinea pig hearts have also shown that after the initial events elicited by scorpion toxin, oscillations in heart rate, contractile force and coronary flow are observed during a period of 5-15 min. Recording of the electrical activity of the heart shows that the oscillations of rhythm are due to wandering pacemakers; as atropine prevents the appearance of such pacemakers it seems likely that they are related to release of acetylcholine by toxin[1].

Our group has also performed experiments with toxin, the most important toxin extracted from *T. serrulatus* scorpion venom[41,43,48]. Injection of toxin in isolated guinea pig heart evokes complex effects, which could be divided into 3 phases: an initial phase (tachycardia or bradycardia associated with an increase in contractile force), an intermediate phase (oscillations of the cardiac rhythm) and a third phase (sinus tachycardia). The bradycardia and oscillations of rhythm are cholinergic in nature, whereas the tachycardia and the increase in contractile force are adrenergic in nature[45,46].

According to some authors[32,33] there is no value of antivenom in the treatment or prevention of the cardiovascular manifestations of scorpion poisoning. Our group does not agree with this statement, based on laboratory and clinical evidences[6,7,20,21,24].

Recently, we have shown that perfusion of the isolated guinea pig heart with Locke solution containing 2% of Butantan antivenom prevents almost totally the cardiovascular effects induced by crude *Tityus serrulatus* scorpion venom[45,46].

The treatment of severe cases of scorpion poisoning should be performed in an Intensive Care Unit, consisting of symptomatic measures, support of vital functions and neutralization of circulating venom[6,7,21].

Pulmonary edema. Many patients stung by scorpions of several species die with pulmonary edema[21]. It seems that the lung edema would be due, at least in part, to a heart failure, caused by an increase in afterload,[2,27] a decrease of left ventricular compliance,[34] the presence of a severe sinus tachycardia[10,21] a myocardial damage[40] and an increase of venous return (preload), caused by discharge of catecholamines[38,44]. Another mechanism contributing to the genesis of pulmonary edema, following the toxin injection, would be the release of substances which increase the vascular permeability, such as kinins, histamine and prostaglandins[5,27,42].

Respiratory effects. Injections of venoms from several scorpion species produce complex respiratory arrhythmias, such as tachypnea, hyperpnea, periodic respiration and apnea[21] Experiments performed by our group have shown that bilateral cervical vagotomy and denervation of carotid bodies prevent the gasping, ataxic

and periodic respiration induced by the toxin[22]. Moreover, local anesthesia of the cervical vagus nerves with lidocaine abolishes the apnea induced by the toxin[24]. These experiments seem to indicate that the respiratory arrhythmias evoked by scorpion toxin are reflex in nature. To test such a hypothesis our group recorded the action potentials of the vagus and phrenic nerves, simultaneously. The apnea evoked by scorpion toxin was accompanied by suppression of phrenic nerve activity, while the action potentials in the vagus nerves were increased in amplitude and/or frequency. Anesthesia of the vagus nerves with lidocaine or bilateral vagotomy suppressed the vagus nerve potentials which were present during the apnea and reestablished the phrenic nerve activity. Based on these data, we have concluded that the apnea elicited by scorpion toxin is due to a reflex mechanism, evoked by stimulation of vagal afferent fibers[21].

Effects on the neuromuscular junction. Injection of *Tityus serrulatus* scorpion venom induces the release of an acetylcholine-like substance from the inervated but not from the denervated rat diaphragm[49]. The release of this substance could account for the twitches observed and also for the decurarizing activity of scorpion venom[14]. On the other hand, electrophysiological studies on the action of tityustoxin, purified from the venom of *Tityus serrulatus*, on rat phrenic nerve-diaphragm muscle preparation, have shown that the toxin has a presynaptic and postsynaptic action at the neuromuscular junction[50].

Effects on the gastrointestinal system. An intense sialagogue effect, induced by *T. serrulatus* scorpion venom in mice, is abolished by atropine, being, therefore, cholinergic in nature[15]. Our group has studied the effect induced by a purified scorpion toxin in rats. We have shown that the toxin increases the flow, the kallikrein and amylase secretions. Pharmacological tests have shown that the increased salivary secretion induced by scorpion toxin in rats is due to cholinergic and adrenergic mechanisms[8].

Our group has shown that an intravenous injection of a toxin obtained from *T. serrulatus* scorpion venom, in anesthetized rats, evokes a dramatic increase in volume, acid and pepsin output of gastric juice and a significant decrease in its pH. Acute bilateral cervical or abdominal vagotomy does not prevent the gastric secretion elicited by toxin, whereas atropine or cimetidine abolish partially or totally thc toxin effects[12,31]. Recently, we have shown that scorpion toxin induces the release of acetycholine and histamine from rat stomach[13]. Based on these results, we think that scorpion toxin exerts its action upon the rat stomach by releasing acetylcholine directly from its nerve endings stores and by releasing histamine directly or indirectly from histaminocytes. Release of gastrin from phyloric antrum could also contribute to the overall response *in vivo* (Cunha-Melo, personal communication).

Injection of *Tityus trinitatis* scorpion venom induces an increase in both volume and amylase secretion from the pancreas of anesthetized dogs[3]. Our group has studied the effects of a toxin obtained from *Tityus serrulatus* on pancreatic secretion, in anesthetized rats. The intravenous injection of toxin causes a striking increase in flow rate, protein content, kallikrein and amylase activities of the pancreatic juice. Sub-diaphragmatic bilateral vagotomy does not prevent the pancreatic secretion induced by toxin, but pre-treatment of the rats with atropine blocks the secretion evoked by toxin. We have presented the hypothesis that the pancreatic secretion induced by scorpion toxin is due to actions of acetylcholine, released from postganglionic nerve fibers, on muscarinic receptors[39]. This hypothesis is supported by *in vitro* experiments[28]. Venoms from scorpions of the genus *Tityus* induce acute pancreatitis in laboratory animals[4,37].

Incubation of segments of guinea pig ileum with *Tityus serrulatus* scorpion venom releases an acetylcholine-like substance which would be responsible by the contraction of the ileum evoked by the venom[16]. However, our group has shown that atropine does not prevent totally the contraction evoked by the toxin either in rat or guinea pig ileum.[11,26] But we have also shown that the toxin releases acetylcholine from rat and guinea pig ileum, through stimulation of sodium channels[18,26]. We have postulated, then, that the contraction of intestinal smooth muscle elicited by scorpion toxin is due to release of acetylcholine and another chemical mediator. Based on indirect evidences we presented the hypothesis that this second mediator could be substance P[11].

Release of noradrenaline and acetylcholine by scorpion toxins. Many investigators have shown that *Tityus serrulatus* scorpion venom or one of its purified toxins (tityustoxin) release noradrenaline and acetylcholine in several preparations, and that blockade of sodium channels with tetrodotoxin prevents their release[17,30,47]. It seems likely that scorpion toxin binds to a site different from the tetrodotoxin-binding site, but its physiological role (e.g. release of acetylcholine) depends on an action on sodium-conducting pore of the channel[9,18,26]

RESUMO: Os resultados apresentados nesta conferência parecem indicar que os efeitos induzidos pelas toxinas de escorpião, obtidas do veneno de *Tityus serrulatus*, são devidos a ações em sítios específicos dos canais de sódio, com despolarização subseqüente das membranas. As arritmias respiratórias são devidas, pelo menos em parte, a despolarizações de fibras aferentes viscerais, induzidas pela toxina, enquanto que os outros efeitos descritos são devidos principalmente a liberação, pelas terminações nervosas, de mediadores químicos como a acetilcolina e catecolaminas.

UNITERMOS: Veneno de escorpião; farmacologia.

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