Chapter V

Discussion

The effect of the intrarenal arterial infusion of a small dose (0.02 mg/kg) of Russell's viper venom on general circulation in intact animals showed a transient fall in mean arterial blood pressure (MAP), narrow pulse pressure (PP) and a decrease of heart rate (HR). These results were similar to those of previous studies in dogs given 0.1 mg/kg of venom intravenously (Tungthanathanich et al., 1986; Tongvongchai, 1984). The changes in mean arterial blood pressure,pulse pressure and heart rate during the first 30 minutes period has been suggested to be due to the vasovagal effect (Chopra and Chowhan, 1934; Lee and Lee, 1979), which could be prevented by vagotomy (Lee and Lee, 1979). Vick and co-worker (1967) proposed that vasodilation and pooling of blood in the hepato-splanchnic area during envenomation and the hypotension was prevented by evisceration. However, this suggestion may not apply to the present results, because a marked decrease in blood pressure and cardiac output were still apparent after envenomation in either splenectomized (Tongvongchai, 1984) or intravascular volume expansion with dextran solution in dogs (Chaiyasest, 1985). The hypotensive action of Russell's viper venom may partly be due to prostacyclin (PGI_2) release, cause vasodilation in the periphery, combine with the release of thromboxane A_2 (TXA_2), leukotriene and also histamine from lungs, cause increase lung perfusion pressure, which induce greater hypotensive effect (Huang,

1984_a; 1984_b). Moreover, Huang and Lee, (1985) believed that the vasodilators relaxed vascular smooth muscle by mechanism involving cyclic-GMP and lipoxygenase products. In the present studies, the rise in mean arterial blood pressure and narrow pulse pressure following the transient decrease after envenomation may relate to catecholamines release as a compensatory mechanism. This view could be supported by the increase in systemic vascular resistance and packed cell volume (Chaiyabutr et al., 1984). The degree of an increase of mean arterial blood pressure and narrowing of pulse pressure in TPTX animals after venom infusion were less than intact animals would be to relate with a low level of extracellular calcium. Since, a low extracellular calcium ion concentration could attribute to contractility of the ventricular myocardium and vascular smooth muscle (Maxwell et al., 1963; Haddy et al., 1963; Chomdej et al., 1977; Stulz et al., 1979; Andersen et al., 1984). Moreover, the release of catecholamine has been known to be calcium dependent (Rubin, 1970). Therefore, the decrement of extracellular calcium in TPTX animals may decrease the release of catecholamines from adrenal medulla (Douglas and Rubin, 1961; Greenberg and Kolen, 1966) and transmitter from adrenergic nerve endings (Kirpekar and Misu, 1967; Boullin, 1967).

The effect on renal hemodynamics of Russell's viper venom in the present study (table 3,4) demonstrated that the venom produced a marked decrease in renal hemodynamics during the initial venom administration in the intact animal. The significant increase in mean arterial blood pressure following envenomation with a parallel increase in renal vascular resistance throughout the period

of experiments indicate a local vasoconstrictor in the kidney (Chaiyabutr, 1985). This effect will cause decrease in effective renal plasma flow, effective renal blood flow and glomerular filtration rate. The decrease toward normal range after the transient rise in filtration fraction after envenomation shows that the decrease in glomerular filtration rate and effective renal plasma flow are in equal proportion (Chomdej and Navar, 1979).

The mechanisms of renal hormone interaction involved in intrarenal vasoconstriction may relate to renin-angiotensin system (Chaiyabutr et al., 1985) and/or prostaglandins (Tongvongchai, 1984). In the present study in TPTX which exhibited hypocalcemia may alleviate the hemodynamic effect of Russell's viper venom which recovered to the control level in a short time as compared to intact animals. A possible endogenous mechanism for releasing and/or action of the hormones induce vasoconstriction after envenomation may be due to calcium dependent (Kirpekar and Misu, 1967; Rasmussen et al., 1986; Henrich and Campbell, 1986). Calcium ion may be direct required for regulation of vascular smooth muscle tone (Blaustein, 1977). Since the intracellular calcium ion supply is necessary for the messenger functions of cyclic-AMP in the action of peptide and amine hormones (Rasmussen et al., 1986) and for activation of the contractile elements has been shown to be critically dependent upon extracellular sources (Stulz et al., 1979).

It has been generally accepted that acidosis results in hyperkalemia because of shifts of potassium from the intracellular compartment for keeping the intracellular to extracellular hydrogen ion and potassium ion ratios equal. Therefore, alteration of the

hydrogen ion ratio leads automatically to a change in serum potassium (Fenn and Asano, 1956; Perez et al., 1981). In the present study, an increase in the plasma potassium concentration is not only consistent with above mentioned. An explanation for this might be that alpha and beta adrenergic receptor stimulations by catecholamine, causes potassium release into the extracellular fluid (Craig and Mendell, 1959). The marked decrease in renal excretion of potassium might also contribute to the sudden increased in plasma potassium concentration (Cox, 1981). These results suggest that renin-angiotensin aldosterone system might be activated after venom infusion caused by the increase in plasma potassium concentration and the reduction of renal blood flow which enhance the formation of angiotensin (Cox., 1981 Blair-West et al., 1962). In the present results, an increase in renal potassium excretion might not only attribute to the filtered load. It should be a characteristic of the aldosterone action which caused to increase in fractional excretion of potassium accompany with a decrease in fractional excretion of sodium and chloride. The increase of plasma chloride concentration may be due to the decrease in urinary excretion and intravascular hemolysis because of red blood cell is rich of chloride (Cohen, 1977). In the present study, free water clearance gradually decreased following the slight increase after envenomation which corresponding to the increase in osmolar clearance of both groups. These results indicate that renal tubular activity of both groups of animals have a normal functions to concentrate urine during the experimental period. However, the increase in arginine vasopressin release, most likely as the consequence of hyperosmolarity, might be contributed to an

increase in water reabsorption and systemic vasoconstriction after venom administration as seen in glycerol induced acute renal failure (Hofbauer et al., 1977; 1982). It has been well known that tubular necrosis and acute renal failure manifested in experiment and human victim following viperine envenomation (Sitprija et al., 1976; Jeyarajahah, 1984; Retcliffe and Pukrittayakamee, 1985). These manifestations might not be expected in the present study. Such difference could be partly due to the difference in dose of administration, species differences in dose tolerance and duration of the study.

However, the main object of this study was to determine the phosphaturic effect of Russell's viper venom. During intrarenal arterial infusion of the venom, an increase in fractional excretion of inorganic phosphorus (FE_{Pi}) was apparent in both the presence and absence of endogenous parathyroid hormone (PTH). The magnitude of the phosphaturic response in acutely TPTX dogs increased significantly, while the increase was not significant in intact dogs. It has been reported that PTH (Schneider et al., 1975) and hypocalcemia (Popovtzer et al., 1975) are responsible for the enhanced $U_{Pi}V$ and FE_{Pi}. The phosphaturic effect in the absence of endogenous PTH clearly indicates a PTH independent action of Russell's viper venom. The decrease in plasma calcium concentration observed in TPTX animals throughout the experiment does not adequately explain the phosphaturia following venom infusion. It could be excluded the effect of volume expansion in Russell's viper venom inducing phosphaturia by a constant of packed cell volume following envenomation in both groups.

Possible disturbance in acid-base balance could not be ruled out

in the increase in urinary Pi excretion following venom infusion, since the urinary titratable acid excretion (U_HV) increased significantly in TPTX group whereas it is not significantly increased in the intact group. An increase in urinary inorganic phosphorus excretion was significant correlation with the U_HV in the post envenomation period (intact group $r = 0.933$; TPTX group $r = 0.958$, $P<0.01$) (Fig 30). This relationship has also been reported in a wide variety of renal diseases in human, but the increase in U_HV was secondary to the increase in PiV (Gyory et al., 1968). Many factors has been reported to influence Pi excretion, eg., PTH, acidosis, C-AMP and extracellular volume expansion. However, factors affecting sodium transport could affect Pi excretion (Dousa and Kempson, 1982). Thus, the increase in FE_{Pi} after venom infusion might not be solely caused by acidosis. After venom infusion the inhibitory effect of venom may not be limited to the transport of Pi, since the increase in fractional excretion of sodium (FE_{Na}) along with FE_{Pi} was observed inboth groups. It is posible that Russell's viper venom affects Na-Pi coupled transport along the bruch border membrane of proximal tubule, as same as aluminum action (Dousa and Kempson, 1982). The sodium dependent transport of fluid, Pi and glucose in the proximal tubule can be inhibited with metabolic inhibitors by blocking mitochondrial ATP production (Gullans et al., 1982). There is evidence that Russell's Viper Venom acts as metabolic inhibitor in the kidney (Chyabutr et al., 1985) .In the present study whether Russell's viper venom act as a mitochondrial inhibitor in the kidney or interact directly with specific site that control the transport of ion across tubular cell remain to be elucidated.

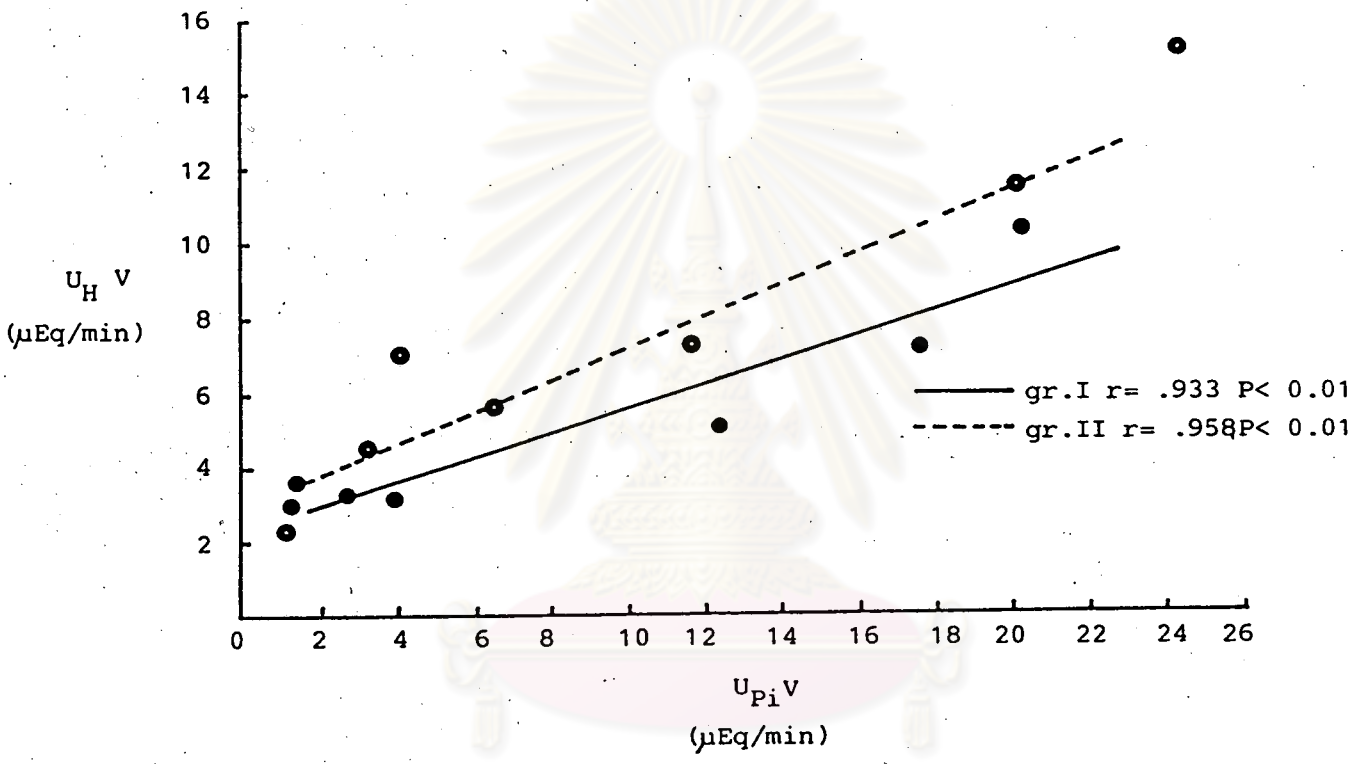

Figure 30 : The relationship between urinary excretion of inorganic phosphorus ($U_{Pi}V$) and the urinary excretion of titratable acid (U_H V) after intrarenal arterial infusion of Russell's viper venom.The solid line is the regression line for intact animals (gr.I).The interrupted line represent the regression line for TPTX animals (gr.II).

In the present study, it dose not know whether the venom activates the adenyl cyclase-cyclic AMP system as some bacterial toxin (Drazin et al., 1971; Moss et al., 1976; Heyningen, 1976) or inhibits the enzyme that destroys c-AMP (Richards et al., 1965; Kocholaty et al., 1971; Stolc, 1985). Recently, it has been evidenced that viperine venom enhances the c-AMP content in human mononuclear and polymorphonuclear leukocytes (Stolc, 1985). Unfourtunately, urinary excretion of c-AMP during pre and post envenomation period can not be measured in present study. However, it is possible to suggest that Russell's viper venom affects tubular reabsorption of Pi may be partly due to the enhance of tubular cell c-AMP production, the inhibitor of Na-Pi cotransport system.

The marked pallor of the surface of the left envenomated kidney in the intact animals may also be interpreted as reflecting a state of cortical hypoperfusion. It seems most likely that the cortical ischemia and the reduced glomerular filtration rate per nephron are responsible for both the oliguria and the reduction of fractional excretion of electrolytes. Because of the marked decrease in the filtration rate per nephron, the electrolyte load which delivered to the tubules may be well below the maximum reabsorption capacity leading to an enhance reabsorption of the filtered electrolyte as seen in mercury induced oliguric acute renal failure (Popovtzer et al., 1971). The significant increased of fractional excretion of electrolyte in TPTX animals could be observed as in nonoliguric acute renal failure induced by mecuric chloride, which could not be explained by secondary hyperparathyroidism, acidosis, extracellular volume expansion or hypocalcemia per se.

In conclusion, these results may indicate that the intrarenal arterial infusion of Russell's viper venom has a direct inhibitory effect on tubular Na-Pi coupled transport. The possible mechanism may be mediated by either, enhanced c-AMP content or act as metabolic inhibitor in the renal tubular cell, which cause alteration of the specific site that control sodium and inorganic phosphorus transport. It is unlikely that parathyroid hormone, hypocalcemia or extracellular volume expansion, factors known to increase urinary Pi excretion, were responsible for the enhanced fractional excretion of Pi after envenomation. The disturbance in acid-base balance could not be ruled out during envenomation. The changes of general circulation, renal hemodynamics and tubular function in TPTX dogs are alleviated as compared to the intact dogs. It is possible that, extracellular calcium concentration contributed to the sequence of intracellular events which culminate in those responses.