RESEARCH PAPER

Chlorpheniramine attenuated hypertensive effect of phenylpropanolamine in rats

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Abstract

Phenylpropanolamine (PPA) is a sympathomimetic drug that exerts its effect mainly through the alpha-adrenergic receptor. A pharmacoepidemiological study reported by the hemorrhagic stroke project in the year 2000 suggested an association between high dose PPA and hemorrhagic stroke. However, the hemodynamic effect of PPA might be altered in the presence of chlorpheniramine (CPM) as used in over-the-counter cold remedy (PPA, 0.3 mg/kg plus CPM, 0.04 mg/kg). It was found that PPA (5-30 times that used in humans) dose-dependently increased in blood pressure in rats and this effect was antagonized by CPM in male rats and non-proestrous female rats but not in proestrous female rats. The hypertensive effect of PPA could not be antagonized by loratadine, a highly specific H1 blocker.

Introduction

Phenylpropanolamine (PPA), a sympathomimetic drug, acts directly on alpha, and also, to a lesser degree, on beta-adrenergic receptor. PPA has an indirect effect by releasing norepinephrine from its storage sites (Hardman et al, 2002, Katzung, 2001). It is most frequently used alone and in combination in various over-the-counter (OTC) cold remedy products as a nasal decongestant. PPA alone in a high dose is also used as weight control drug (Hardman et al, 2002, Katzung, 2001).

Although adverse drug reactions and interactions are continuing to be documented in the literatures, the safety of PPA remains controversial. Serious side effects such as hypertensive crisis (Humber-stone, 1969, Gibson and Warrell, 1972), Horowitz et al, 1980, Lake et al, 1980, Thomas et al, 1991, Hantsch et al, 1997), stroke (Horowitz et al, 1980, Kernan et al, 2000, Kikta et al, 1985, Mersfelder, 2001, Delorio, 2004, Hamilton and Sharieff, 2000), seizure (Delorio, 2004, Hamilton and Sharieff, 2000), and arrhythmia (Pilsczek,

2003) have been reported following consumption of PPA. In 2000, a pharmaco-epidemiological study by the hemorrhagic stroke project reported an association between high dose PPA and hemorrhagic stroke (Kernan et al, 2000). Thus, the US FDA issued public warning on the risks of PPA and requested the OTC manufactures to voluntarily remove PPA from the market (Horwitz et al, 2000, FDA, 2000a, FDA, 2000b). In 2001, Thai FDA, under the Ministry of Public Health, decided to withdraw PPA from the market as it is recognized to be unsafe for OTC use for nasal decongestant or weight control.

In 2005, there was a reappraisal on this issue was introduced as it states that the US FDA's regulatory request launched in year 2000 might have been premature (Stier and Henneken, 2005). In 2012, there is a suggestion to allow PPA, in cold and sinusitis preparations, as registered ethical products to be brought only with prescription and used under medical supervision with watchfulness for the dose and any drug interaction (Yahoot, 2012).

However, no scientific work supports the unsafe information of PPA especially in combination of chlorpheniramine (CPM) which is the most common use in combination with PPA in cold remedies. Pharmacokinetic and hemodynamic effect of PPA was altered in the presence of caffeine and/or CPM tested in rats. Co-administration of PPA with caffeine increases the blood level of PPA by 30% while CPM decreases PPA by 15%. Caffeine co-administered with CPM and PPA caused 60% increase in PPA levels. On contrary, CPM increases in PPA level in the brain (Kaddoumi et al, 2004).

The purpose of this work was to assess the effect of PPA on blood pressure when administered alone or co-administered with CPM and to evaluate possible mechanism of drug interaction on hemodynamic effect.

Materials and methods

Animals: Male and female Wistar rats (200-250 g) were obtained from The National Laboratory Animal Center, Mahidol University. The animals were acclimatized in groups of two or three per cage in a temperature controlled room (25±1°C) for at least one week before the experiment. Standard chow and tap water were supplied *ad libitum*. The experiment protocol was approved by the Institutional Animal Care and Use Committee, Faculty of Pharmacy, Mahidol University, Thailand (approval no. PY0004).

The drugs used in the present study were phenylpropanolamine HCl (PPA, ALPS Pharm, Japan), chlorpheniramine maleate (CPM, Venkatarama Chem, India), loratadine (Cadila Pharma, India). All drugs were freshly dissolved in distilled water before the experiment.

Effect of phenylpropanolamine on blood pressure in male rats: The systolic blood pressure (SBP) were determined by indirect blood pressure measurement at caudal artery in conscious rats using blood pressure recorder (Ugo Basile,Type 8006, Italy).

Each rat was trained, for 1 week prior to test, with the tail cuff method by restricting its movement in a rat restrainer and giving the tail cuff pressure so as to get its acquaintance to the procedure. Before experimentation, each rat was placed under two 100 watt lamps for about 30 min until obtaining sufficient vasodilatation of caudal artery, then, it was placed in the restrainer for 3 min to reduce the stresses from warming. PPA at doses

of 0.3, 1.5, 4.5, 9.0 mg/kg BW (1, 5, 15, 30 times of human doses), respectively) were orally administered to the rats assigned to be in the experiment group, while water to the control group. The SBP were measured at initial (before administration) and at 30, 60, 90 and 120 min after administration.

Effect of co-administration of PPA and CPM on SBP: The same experiment protocol was used. PPA (9.0 mg/kg BW) and PPA in combination with 0.04, 0.12, 0.24 mg/kg BW (1, 3, 6 times of human doses, respectively) of CPM were orally administrated to the male rats.

Effect of estrous cycle on SBP and HR after feeding by PPA alone and in combination with CPM in female rats: The same experiment protocol was used. PPA (9.0 mg/kg BW) alone or combined with CPM 0.04, 0.12, 0.24 mg/kg BW (1, 3, 6 times of human doses, respectively) were orally administrated to female rats both proestrous and non proestrous cycle.

Effect of phenylpropanolamine combined with loratadine on blood pressure: The same experiment protocol was used. PPA (9.0 mg/kg BW) alone or combined with loratadine 0.2, 0.6, 0.12 mg/kg BW (1, 3, 6 times of human doses, respectively) were orally administration to male rats. The SBP were measured at initial (before drug administration) and at 30, 60, 90 and 120 minutes after drug administration.

Statistical Analysis: Restuls were expressed as mean ± the standard error of mean (S.E.M). Data from treated animals were compared with those of normal and the hypertensive control groups. Statistical significances between the treated groups and the respective control groups were determined by one way analysis of variance (ANOVA) followed by the multiple comparison test of the Post Hoc test using the program SPSS 10.0 for Windows. Statistical differences with $p < 0.05$ were considered significant.

Results and Discussion

Fig. 1 demonstrates that oral administration of PPA increases SBP of the rats. This effect was observed from 30 min and lasted for at least 2 h. At a single dose, which is equivalent to a human dose of 0.3 mg/kg, PPA slightly increased SBP of the male rats. However, at higher doses, for example, 5 times higher than that used in human of 1.5 mg/kg of PPA could significantly increase SBP of the male rats at 30 and 60 min ($p<0.01$). PPA at the dose of 4.5 mg/kg, about 15 times of the human dose also gave a significant increase in SBP of the male rats at 30, 60 and 90 min ($p<0.01$).

The highest dose of PPA used in the present study at 9.0 mg/kg which was 30 times of the human dose significantly increased SBP of the rats for the entire duration of study from 30 to 120 min. This finding suggests the hypertensive effect of PPA in a dose dependent manner.

Fig. 2 showed that CPM, at the doses of 0.12 and 0.24 mg/kg, which were 3 and 6 times equivalent to the human dose, respectively, co-administered with PPA at a dose of 9.0 mg/kg (30 times of the human dose) could suppress the hypertensive effect of PPA, in comparison with the control PPA treated group. CPM at the dose of 0.04 mg/kg did not show such effect in rats.

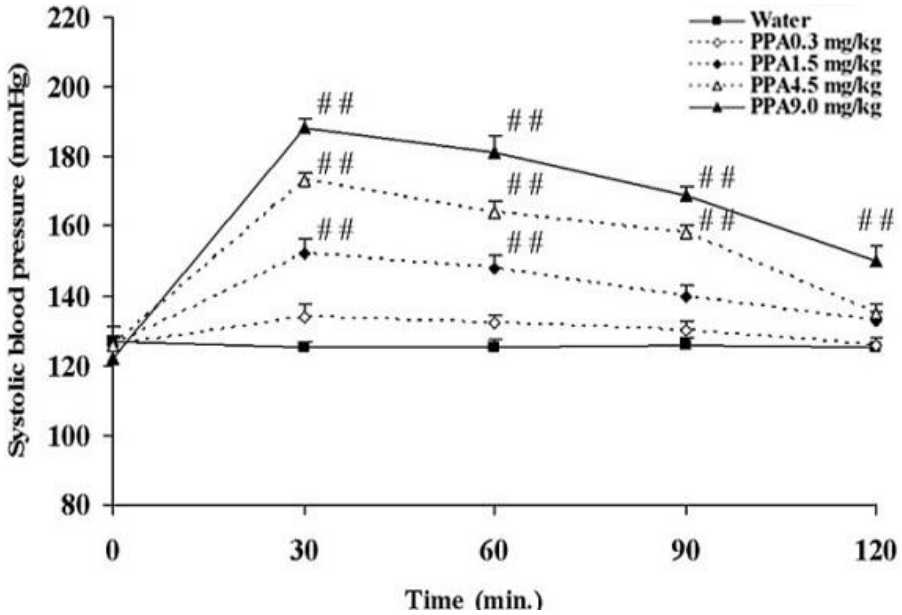


Figure 1 Effect of PPA (1.5, 4.5 and 9.0 mg/kg) on systolic blood pressure in rats. ##P<0.01 when compared with the corres-ponding normal control value. Data are mean ±SEM of 10 rats each.

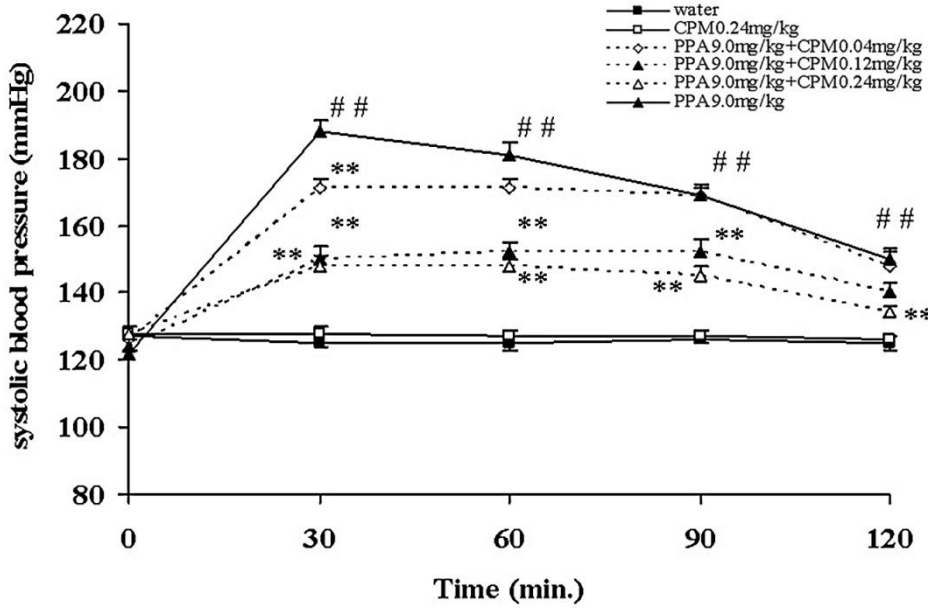


Figure 2 Effect of PPA (9.0 mg/kg) combined with CPM (0.04, 0.12 and 0.24 mg/kg) on systolic blood pressure in male rats. $^{\#\#}$P<0.01, **P<0.01 when compared with the corresponding normal control and hypertensive values, respectively. Data are mean ±SEM of 10 rats each.

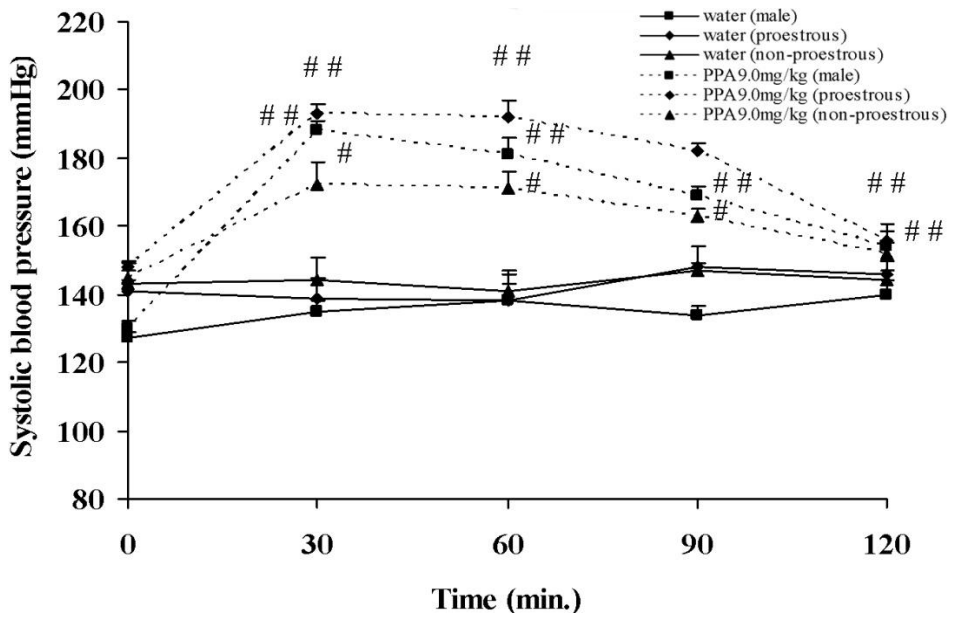
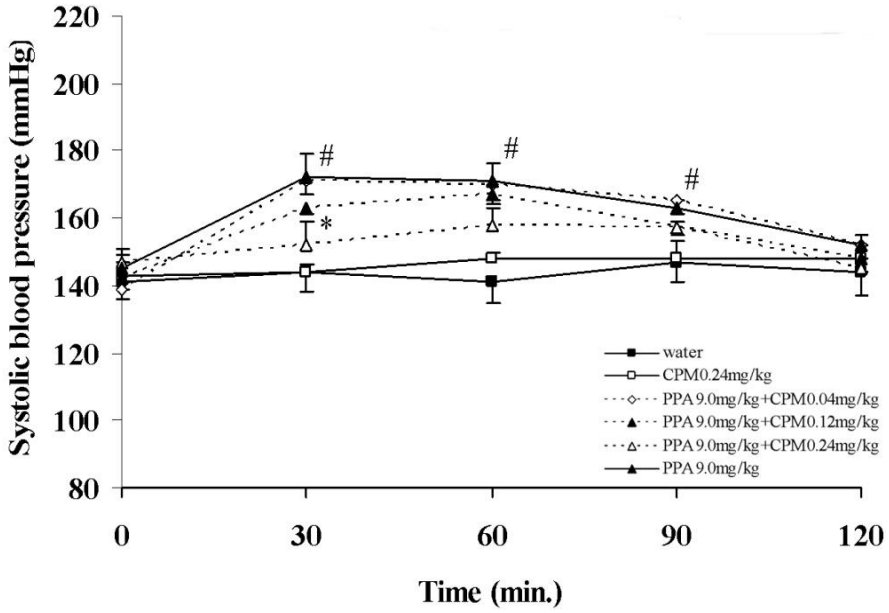


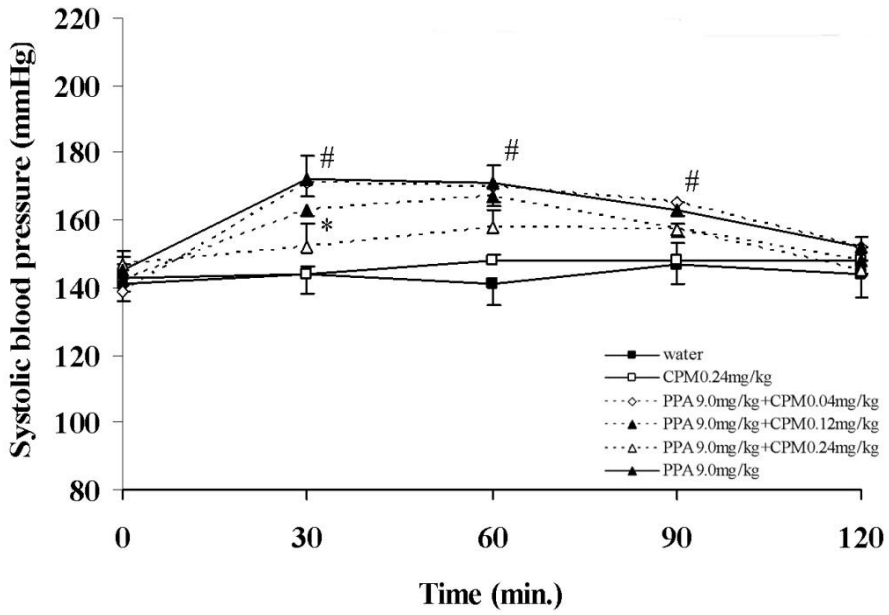
Figure 3 Effect of sexes and oestrous status on systolic blood pressure of rats administered single oral doses of PPA of 9.0 mg/kg BW. (mean ± S.E.M, n = 6) # representing $p < 0.05$ and ## $p < 0.01$ from the respective control group (water) at the corresponding time

In comparison of SBP among proestrous female rats, non-proestrous female rats, and male rats treated with PPA at the dose of 9.0 mg/kg BW, SBP of the proestrous rats was highest during the duration of study, as shown in Fig.3.. Non-proestrous rats showed lowest SBP among the PPA administration groups. Proestrous female rats which were co-administered with CPM (0.04, 0.12, and 0.24 mg/kg BW) could not reduce hypertensive effect of PPA, as shown in Fig. 4 (a), while in non-proestrous female rats, CPM reduced hypertensive effect of PPA in a dose response manner, as shown in Fig. 4 (b). These findings suggested that CPM attenuated the hypertensive effect of PPA in a dose dependent manner. The mechanism of attenuating effect of PPA by CPM may involve various activities due to several pharmacological actions of CPM including H_1 blocker, serotonin blocking actions, anti-cholinoceptor actions and adrenoceptor-blocking actions etc. (Katzung, 2001). These pleiotropic actions of CPM might represent the possible mechanisms of CPM in attenuating the hypertensive effect of PPA.

Loratadine (a histamine H_1 receptors blocker) was attempted in order to explain the antihistamine effect of CPM on attenuating hypertensive effect of PPA. Results in Fig. 5 show negative effect of loratadine on hypertensive reduction of PPA (Fig. 5). Thus, antihistaminergic action of CPM migh not affect PPA in this aspect.



(a)



(b)

Figure 4 Effect of PPA (9.0 mg/kg) combined with CPM (0.04, 0.12 and 0.24 mg/kg) on systolic blood pressure in (a) proestrous female rats and (b) non-proestrous female rats. $^{\#\ \#}P<0.05$, $^{*}P<0.05$ when compared with the corresponding normal control and hypertensive values, respectively. Data are mean ±SEM of 10 rats each.

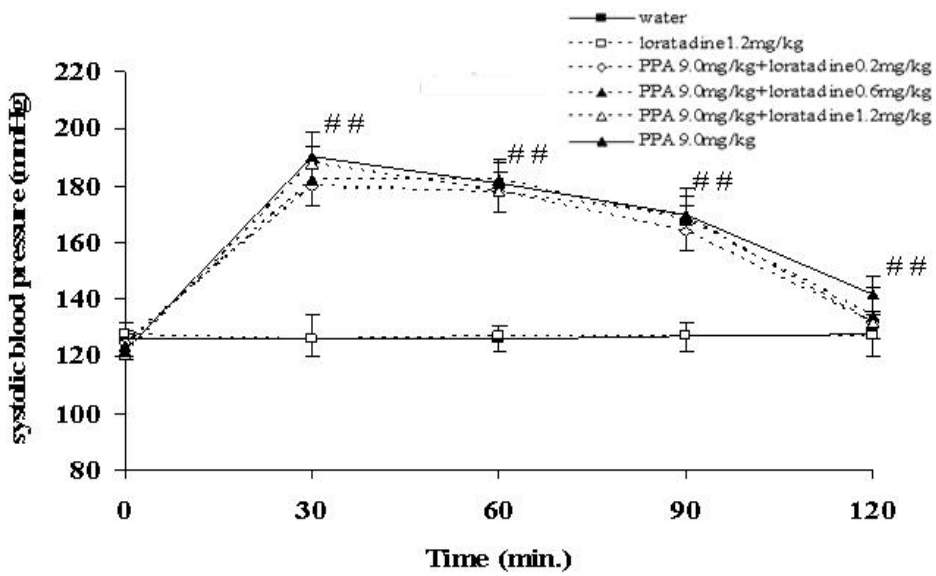


Figure 5 Effect of PPA (9.0 mg/kg) co-administered with loratadine (0.2, 0.6 and 1.2 mg/kg) on systolic blood pressure in rats. $^{\#\#}P<0.01$ when compared with the corresponding normal control value. Data are mean ±SEM of 6 rats each.

The serious side effect of PPA, hypertension, have been reported in patients following consumption of PPA. In consistence with the previous reports, our study showed that PPA had hypertensive effect in a dose dependent manner. PPA at the doses of 0.3, 1.5, 4.5 and 9.0 mg/kg BW (1, 5, 15, 30 times of the human dose, respectively) significantly increased SBPs in a dose response manner. As there were few reports of hypertensive effects of PPA when using in a combination with CPM as a cold remedy, we designed an experiment in order to investigate the possible interaction of this two drugs on blood pressure in rats because the most common use of PPA in the markets was a low dose of PPA combined with CPM as a cold remedy drug. Therefore, an experiment was conducted in order to study the effect of PPA combined with CPM on blood pressure. The result showed that CPM significantly attenuated the hypertensive effect of PPA in a dose dependent relationship in male rats (Fig. 2). In female rats, PPA in combination with CPM, CPM could reduce hypertensive effect of PPA in a dose response manner in non-proestrous group but not in proestrous female rats. Since during the proestrous stage, the level of estogen is higher than that of non-proestrous stage (Takezawa et al, 1994), therefore estrogen may involve in the potentiation of the hypertensive effect of PPA which could not reduce by CPM. The mechanism of this effect of estrogen has not been fully identified. This data supported the Yale's study that the hypertension and stroke induced by PPA often occurred in women more than in men (Horwitz et al, 2000).Since our study showed that CPM could attenuate the hypertensive effect of PPA, it was interesting to find the mechanism of attenuating effect on PPA by CPM. As CPM has various pharmacological actions i.e. H1 blocker, serotonin blocking actions, anti-cholinergic actions and adrenoceptor-blocking actions, etc (Katzung, 2001), one or several of these pleiotopic actions of CPM might be the possible mechanisms that attenuate the hypertensive effect of PPA. Loratadine

(H1 receptors blocker) was used to test the antihistamine effect of CPM in attenuating the hypertensive effect of PPA. It was found that loratadine did not reduce the hypertensive effect of PPA. Therefore, anti-histaminergic action of CPM could be excluded. However it is interesting to further investigate the possible mechanism of action for attenuating the hypertensive effect of PPA by CPM

Conclusion

From the present results, scientific data can be summarized that support the relationship of PPA and blood pressure with in combination CPM in three aspects. First, PPA exerts hypertensive effect in a dose response manner. Second, the hypertensive effect of PPA could be attenuated by CPM in male rats. Third, the hypertensive effect of PPA could not be attenuated by CPM in female in the period with high estrogen.

Acknowledgements

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