INTESTINAL CONTRACTIONS UNDER THE INFLUENCE OF ETHANOL USED FOR SIMPLE AND SUCCUSED DRUG DILUTIONS

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ABSTRACT

Ethanol (Eth) is frequently used both as solvent for many drugs and as a vehicle for many medicinal (i.e., allopathic and homeopathic) mixtures and dilutions. No significant difference between simple and succussed dilutions were found on ileal (intestinal) muscles of rabbit except at 2x and 84% ethanol. Simple dilutions of ethanol have been found to decrease the intestinal rate of contraction immediately after administration. The rate was increased after three minutes of administration except at 95% Eth and 1x, Acetylcholine (ACh) and Adrenaline (Adr) dilution in ethanol. The succussed dilutions of ethanol itself also decreased the rate of contraction immediately (1x) and after three minutes of administration (at 1 and 2x). These results suggest that ethanol inhibits the smooth muscle activity due to a probable reduction in cross bridge cycling. However, this effect of ethanol seems to be more pronounced and long lasting after succussion.

Key words: Succussed dilutions, simple dilutions, ethanol, acetylcholine, adrenaline, ileal smooth muscle

INTRODUCTION

Ethanol is psychoactive drug that is similar to the sedative hypnotic compounds. It is soluble in both water and fat and diffuses easily through the biological membrane. Thus it can be absorbed rapidly and completely from the entire GIT, mostly from the upper intestine having large surface area (Julien, 1996). However, Dinda and Beck, (1982) reported that ethanol depresses the hydrolysis of all peptidases in a dose dependent manner. Further, it inhibits the activity of lactase, sucrase and maltase and stimulates alkaline phosphotase at the brush border of intestine (Dinda et al., 1979).

Preedy et al. (1988) observed the effect of an acute dose of ethanol (75mM/kg-body weight) on protein synthesis in the small intestine of the rats. They found that protein extracted from contractile apparatus showed 40-50% lesser values when this synthesis was inhibited by ethanol. They also observed the functional disturbances in the gut that was exposed by ethanol. These disturbances have been attributed to decrease amount of contractile apparatus due to reduced smooth muscle protein turn over with the use of ethanol. However, moderate alcohol consumption (101 to 300 ml alcohol/week) may protect against severe coronary atherosclerosis as reported by Koichi et al., (1990).

Ethanol also effects the peristaltic contraction of intestine. Nasreen et al. (1983) reported that Eth can decrease the tone transiently and this decrease is followed by sudden increase in it. In homeopathic dilution ethanol constitute as a “vehicle” because it is the only continuum between low and high dilutions. It was proposed (In homeopathic system of medicine) that a physical factor of unknown nature develop in the vehicle itself that is supposed to be responsible for the dilution activity and importance laid on the process of succussion than on serial (simple) dilution (Dutta, 1991). Further it was also proposed that kinetic energy applied to the system, causes the ethanol molecule to be more energetic, potent and therapeutically active. Azeem et al. (1999) and Arifa et al. (1995, 1997, 2000) have also reported the differences in the effects of simple and succussed acetylcholine and adrenaline dilutions (prepared in deionized water) on cardiac and intestinal activity.

In the present study the effect of simple and succussed dilutions of ethanol, acetylcholine and adrenaline (prepared in ethanol and deionized water) were observed on the intestinal tissue due to the importance of ethanol as solvent in homeopathic system.

MATERIALS AND METHODS

Preparation of simple Dilution:

Simple dilution of 1x concentration was prepared by adding 0.15 ml of Absolute Ethanol (95%) in 149.85ml of deionized water to get a volume of 150 ml. Then 15 ml of this simple 1x dilution was again diluted with 135 ml of deionized water to get 150 ml of 2x dilution. This procedure was repeated to get 3 x dilutions. The 84% ethanol
(Eth) was prepared by adding 126 ml of (95%) Eth in 24 ml of deionized water. This dilution of 84% Eth was also used as a solvent for the preparation of Acetylcholine (ACh) 1x and Adrenaline (Adr) 1x dilutions by dissolving 150 mg of drug (ACh and Adr) in 150 ml of 84% Eth. A concentrated dilution of Eth (95%) was also used to observe its effect on intestine. While simple dilution of ACh 1x and Adr 1x prepared by dissolving 150 mg of drug in 150 ml of deionized water.

Preparation of Succussed dilution:

It is similar to the preparation of simple dilution except that in succussed dilution the simple dilution was shaken for two hours in a mechanical shaker.

Isolation and fixation of intestinal strip:

Both sexes of *Oryctolagus cuniculus* rabbit’s intestine were used to test the effect of simple and succussed dilutions. After decapitation of rabbit, its abdomen was opened immediately and a large piece of ileum was removed and washed by flushing the Kreb’s solution through its lumen. About 2.5 to 3.0cm length of ileum strip was mounted in the organ bath assembly having oxygenated Kreb’s in its gut tube maintained at 37±1°C. First of all, rhythmic activity was isotonically recorded as control by using auxotonic transducer on minigraph (Lafayette instrument company USA). Later, the effect of dilutions was recorded by adding 1 ml of diluted drug directly into the gut bath having 50 ml of Kreb’s. Later, continuous recording was obtained that was later used for the measurement of the rate of contraction per minute. Same procedure was applied for each dilution and pure ethanol. The effect of each dilution was calculated as percent of control. Statistical analysis was done using standard procedures and student’s t-Test for comparison between simple and succussed dilutions.

RESULTS

Effect of Simple Dilutions:

The result of simple dilution presented in Table 1 demonstrates that Eth decreases the rate of contraction immediately after its administration at 1x, 3x and 84% Eth dilutions. While after three minutes, it increased slightly at 1x, 2x, 3x and 84% of Eth. The rate of contraction also reduced immediately by simple ACh 1x and Adr 1x prepared in Ethanol. It later reduced further after three minutes of their administration. However, the response of 95% Eth (both immediately and after three minutes) was either irregular or had no response, respectively.

Effect of Succussed Dilutions:

The result regarding succussed dilutions presented in Table 1 shows that the rate of contraction decreased immediately at 1x, 84% and 95% Ethanol. While, contraction rate slightly increased at Eth 2x and 3x dilutions. Later, after three minutes, 2x, 84% Eth and 95% Eth administration this rate of contraction remained decreased but higher at Eth 1x and 3x dilutions. However, this increase in the rate of contraction at Eth 1x dilution was still lesser than its control. Moreover, succussed Eth+ACh 1x and Eth+Adr 1x dilutions had greatly decreased the intestinal rate of contraction both immediately and after three minutes of administration. However, this effect was so intense at Eth +Adr 1x dilution, that it abolished this intestinal activity completely with no response at all.

A statistical comparison between simple and succussed dilution of Eth 2x showed a significant (P<0.05) decrease after three minutes of administration. Similarly 84% Eth also showed significant decrease both immediately and after three minutes of administration being P<0.05 and P<0.0005 respectively.

Comparison of the effects of dilutions prepared in deionized water Vs in Ethanol:

The results presented in Table 2 demonstrated that simple ACh 1x dilutions prepared in deionized water increased the rate of contraction immediately and later decreased it slightly after three minutes of administration. While simple Ach 1x prepared in ethanol decreased the intestinal rate of contraction both immediately and after three minutes of administration (being greater after three minutes). Moreover, simple Eth 1x dilution prepared in deionized water slightly decreased it but later, increased it after three minutes of its administration.
However, the result regarding simple adrenaline dilutions (Table 2) showed that Adr 1x either prepared in deionized water or in ethanol decreases the intestinal rate of contraction both immediately and after three minutes of administration. However, this reduction was greater in Adr 1x prepared in ethanol (Eth+Adr 1x) as compared to Adr 1x dilution prepared in deionized water.

The result presented in Table 2 regarding succussed dilutions demonstrates that Eth 1x dilution prepared in deionized water slightly decreased (2-5%) the intestinal rate of contraction both immediately and after three minutes of its administration. While the succussed dilution of Ach 1x prepared in deionized water and prepared in ethanol (Eth+ACh 1x) greatly decreased (15-36%) the rate of contraction. However this reduction was found greater in Eth+ACh 1x (prepared in ethanol) when compared to ACh 1x (prepared in deionized water) dilutions.

Similarly, when Adr 1x dilution prepared in deionized water compared with Adr 1x prepared in ethanol it was found that both these dilution also reduced the intestinal rate of contraction but it was decreased tremendously in Eth+Adr 1x upto 80% immediately, while after three minutes it abolished the intestinal activity completely.

DISCUSSION

Simple Dilution:

Decreased rate of intestinal contraction on using simple dilution probably indicate a change in electrical activity (Table 1). According to Sanders and Bauer (1982) and Sanders and Berry (1985) ethanol interfere the excitation contraction mechanism of canine antral muscle and results in a dose dependent reduction in the amplitude and frequency of phasic contraction of circular muscle associated with depressed slow waves in canine antral muscle. Moreover, Puurunen et al. (1977) and Tague and Shanbour (1977) stated that the topical application of ethanol markedly decrease the tissue levels of ATP and other high-energy nucleotide. Further, it also inhibits ATP gated ion channels (Yu et al., 1996). In the present study a decreased ATP concentration due to enhance ATP degradation (Wood et al., 1993) in the presence of ethanol had slowed down the rate of cross bridge cycling per unit time thus, reducing the rate of contraction. While slight increase in intestinal contraction after three minutes of administration of Eth 1x, 2x, 3x and 84% dilutions probably indicate the influence of intrinsic innervation (Paton and AbooZar, 1968; Bauer and Kuriyama, 1982 a, b) that recovers from reduced ATP environment. Eth+ACh 1x reduced intestinal rate of contraction, while ACh alone was found capable to increase it (Table 2). Sanders and Berry, (1985) also reported that phasic contractions are stimulated by ACh but they are inhibited by Eth. While Yu et al., (1996) stated that the ethanol inhibited the neurotransmitter (ACh) gated ion channels by direct interaction with the receptor protein. Thus, in the present study the ethanol inhibited the activity of ACh receptor and produced pronounced inhibitory effect ultimately on cross bridge cycling and rate of contraction.

The effect of Eth+Adr 1x and Adr 1x dilutions demonstrates decreased intestinal contraction and greater inhibitory effect when ethanol containing adrenaline dilution was used (Table 2). According to Hoffman and Lefkowitz, (1991) and Hoffman (1998) adrenaline decreased the frequency of spontaneous contraction by relaxing the activity of alpha and beta-receptor of gut. This relaxation in gut mediated through hyperpolarization and decreased spike activity of smooth muscle. Ethanol also reduces the intestinal activity (Sanders and Berry, 1985) by altering the electrical activity of smooth muscle (Knight et al., 1992). Thus, it is probable that Adr 1x and Eth+Adr 1x had exerted relaxing influence by changing the electrical activity of smooth muscles.

Succussed Dilution:

According to the present results the succussed dilution of ethanol decreased the intestinal rate of contraction except at 2x and 3x dilutions. This decrease in the rate of contraction indicates that its inhibitory action relates with its concentrated dilutions. However, increased rate of contractions at only 3x dilution indicates that the character of drug molecule was probably changed on succussion and thus the rate of contraction was higher. While, the greater reduction observed in all succussed dilutions (except 3x) as compared to their simple one demonstrated that succussion has enhanced the inhibitory response. It may be due to molecular clustering effect in these dilutions as reported by Heintz (1941) and Leeser (1967). Moreover, on the basis of results presented in Table I it is also clear that change in the rate of contraction produced by simple dilutions is not long lasting while, this change produced by succussed dilution persists after three minutes. Such comparative effect probably indicates that succussion is capable to produce molecular change in ethanol that does not allow desensitization of receptors as reported by Bourne (1998), neither it allows intrinsic influence to overcome its response.
CONCLUSION

It is concluded that simple and succussed dilutions of ethanol, ACh (prepared in ethanol) and Adr (prepared in ethanol) differs in their magnitude of effect on intestinal rate of contraction. The effect of succussed dilutions was pronounced and lasted for a longer period than their simple ones, suggesting that succussion resulted in molecular clustering (Heintz 1941; Leeser 1967) that probably enhanced and prolonged their effect, especially for 84% ethanol.

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REFERENCES


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