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HISTAMINE-INDUCED BRONCHOSPASM IN UNANAESTHETIZED GUINEA-PIGS

II. *A Comparison of the Bronchospasmolytic Agents Terbutaline, Orciprenaline and Isoprenaline*

By

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In a pharmacological study of the new bronchospasmolytic agent terbutaline (Bricanyl®) Persson & Olsson (3) presented among other data, also a brief information of the effects on unanaesthetized guinea-pigs in histamine aerosol. The present study reports these results in detail. The ability of terbutaline to prevent histamineinduced bronchospasm is compared with orciprenaline and isoprenaline, both commonly used bronchodilating drugs.

The substances used in the study, besides histamine dihydrochloride, are terbutaline sulphate [1-(3,5-dihydroxyphenyl)-2-(*t*-butyl-amino)-ethanol] (TRB), orciprenaline sulphate (OPR), and ($\pm$) isoprenaline sulphate (IPR). The dose used refers to the base form.

MATERIAL AND METHODS

Male guinea-pigs (200-400 g) were used in the study. According to the description of the histamine aerosol technique (2) a histamine concentration of 0.4 mg/ml (base form) was used in the aerosol solution. The respiratory influence POI-1 (2) was selected as the indication of histamine induced bronchospasm. As mentioned in the methodological report, this stage of spasm appeared within 4 minutes in 96 per cent of untreated (control) animals when the mentioned concentration of histamine was used.

The bronchospasmolytic compounds to be tested were injected subcutaneously (s.c.), intraperitoneally (i.p.), and administrated orally (p.o.) by a stomach

tube to the animals, which were starved for 10 hours but allowed water *ad libitum*. The ED₅₀ of the compound was calculated from the percentage of animals withstanding the histamine mist in > 4 minutes. The time from administration of the bronchospasmolytic compounds to the histamine test was 15 minutes after s.c. and i.p. and 30 minutes after p.o. supply of the drug. The dose 2.0 mg/kg p.o. was given when the compounds were compared for onset and duration of effect. From the group of guinea-pigs given this amount of TRB, OPR or IPR, animals were collected for test in the aerosol 10, 20, 30, 40, 50 minutes and 1, 2, 3, 4, 5, and 6 hours after the oral administration. The percentage of the treated animals withstanding the aerosol > 4 minutes and also > 10 minutes before the stipulated respiratory influence was registered. Control animals were run parallel to test animals and were given 0.8 per cent NaCl. Because the used solution of IPR was stabilized with ascorbic acid (0.2 mg/ml) the saline solution given to corresponding controls also included this stabilizer.

RESULTS

All the used compounds have a protecting effect against histamine-induced bronchospasm in guinea-pigs, and the activity is compared at the dose level where animals are protected more than 4 minutes (ED₅₀). If the activity of the three compounds is compared after s.c. injection (Table 1) the effect of TRB is a bit higher than the one of OPR ($1.2 \times$ OPR), but compared to IPR the effect of TRB is only 0.15. The results obtained after i.p. administration are presented in Table 2. TRB in this part of the study is 4 times more active than OPR, but less than half as active as IPR. After an oral supply of the drugs (Table 3) TRB is about 3 times more active than both OPR and IPR.

If 2.0 mg/kg of the compounds are given p.o. to determine onset and duration of the bronchospasmolytic effect, the results presented in Fig. 1 are obtained. Already 10 minutes after the administration TRB and IPR protect about 75 per cent of the animals > 4 minutes; after about 30 minutes OPR equalled this. OPR has a slow onset compared with TRB and IPR, especially when the intensity of the effect or protection for > 10 minutes is considered. When the duration is compared TRB shows an outstanding effect compared to IPR, and also to OPR.

TABLE 1
Bronchospasmolytic Activity after Subcutaneous Administration.

Compound	ED 50 mg/kg m $\pm$ s.e.
Terbutaline	0.095 $\pm$ 0.007 (110)
Orciprenaline	0.12 $\pm$ 0.05 (70)
Isoprenaline	0.015 $\pm$ 0.003 (50)

The compounds were administered subcutaneously to guinea-pigs 15 minutes before the introduction in histamine aerosol. Dose protecting 50 per cent of the animals > 4 minutes in the aerosol is expressed as the ED 50 of the drug in question. The number of animals used is put in brackets.

TABLE 2
Bronchospasmolytic Activity after Intraperitoneal Administration.

Compound	ED 50 mg/kg m $\pm$ s.e.
Terbutaline	0.15 $\pm$ 0.03 (80)
Orciprenaline	0.60 $\pm$ 0.08 (48)
Isoprenaline	0.065 $\pm$ 0.006 (36)

The compounds were given intraperitoneally 15 minutes before the animals were tested in histamine aerosol. The dose that protects 50 per cent of the animals > 4 minutes in the aerosol is the ED 50 for the drug involved. The number of animals used is put in brackets.

TABLE 3
Bronchospasmolytic Activity after Oral Administration.

Compound	ED 50 mg/kg m $\pm$ s.e.
Terbutaline	0.4 $\pm$ 0.08 (48)
Orciprenaline	1.2 $\pm$ 0.2 (40)
Isoprenaline	1.4 $\pm$ 0.2 (50)

The compounds were administered orally to guinea-pigs 30 minutes before the introduction in histamine aerosol. The dose that protects 50 per cent of the animals > 4 minutes in the histamine is the ED 50 for the drug involved. Number of animals used is put in brackets.

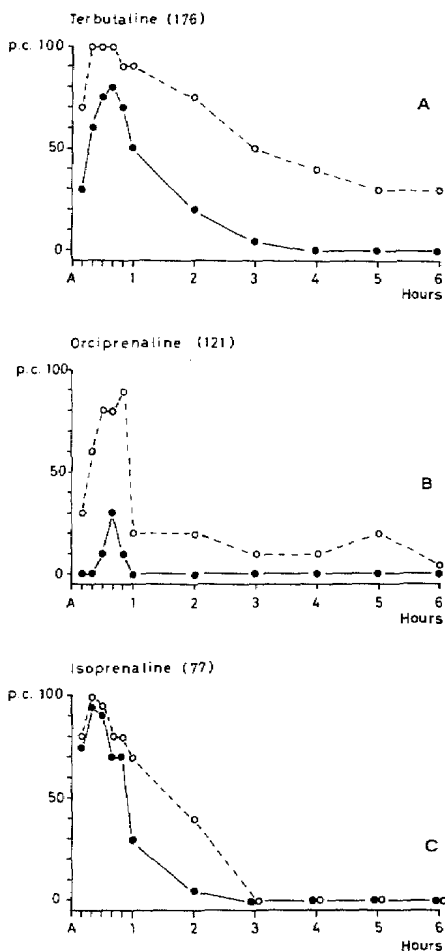


Fig. 1.

The compounds TRB, OPR, and IPR were given orally in a dose of 2 mg/kg to intact guinea-pigs. The animals were tested at different times (abscissa) after the administration (A) with the first determination after 10 minutes and the last after 6 hours. Each animal was tested only once and the percentage (ordinate) protected > 4 minutes (---○---) and > 10 minutes (—●—) in the aerosol was registered. The number of tested animals for each compound is put in brackets.

DISCUSSION

When administered s.c. TRB is more active than OPR in preventing histamine induced bronchospasm, but the difference is not great. IPR is about ten times as active as these two mentioned drugs. The effect of TRB increases compared to OPR when the route of administration is i.p., but still IPR is the most active. The difference in ED₅₀ between TRB and OPR when given orally is of about the same order as after i.p. administration, but after oral administration an obvious decrease of the activity of IPR evaluated as ED₅₀ is noted. In spite of a high initial activity after 2 mg/kg p.o. the duration of the effect is short of this drug.

In the IPR molecule the OH-groups in the benzene ring are placed in 3,4-position. This configuration makes the molecule sensitive to the methylating action of the enzyme catechol-O-methyl-transferase (COMT) (1), and the produced methylated compound lacks bronchospasmolytic activity. This degradation, besides an inactivation by a conjugation, seems to determine the effect, especially after an oral administration. If the OH-groups are placed in the 3,5-position in the ring the mentioned enzymatic action is prevented, and a prolongation of the bronchospasmolytic activity might be obtained. The compound OPR has the OH-groups in this 3,5-position and the same configuration on the amino nitrogen (i-propyl) as in IPR. As predicted, the duration of the effect seems to be prolonged compared with IPR. However, the initial effect of OPR is low, especially after oral administration. In TRB the i-propyl on the nitrogen is changed to t-butyl with the OH-groups still in the 3,5-position. As can be seen from the experiments in this study this latter configuration of the molecule gives a high bronchodilating activity, particularly after oral administration, and in addition a prolonged effect is reached.

SUMMARY

The bronchospasmolytic agents terbutaline (TRB), orciprenaline (OPR) and isoprenaline (IPR) were compared for

ability to protect non-anaesthetized guinea-pigs against histamine-induced bronchospasm. After subcutaneous and intraperitoneal injection IPR was the most effective drug. After oral administration TRB revealed the lowest ED₅₀ and the most prolonged effect.

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