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Absorption of ampicillin from the human lung

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Maddocks, J. L. (1975). *Thorax*, **30**, 68–71. **Absorption of ampicillin from the human lung.** The absorption of ampicillin from the lungs after intratracheal administration was studied in a healthy human. Doses of 50 mg, 250 mg, and 1,250 mg were given by intratracheal injection, and absorption of ampicillin from the lungs was assessed by measuring plasma levels, the area under the plasma level-time curve, and urinary excretion. All these indices of absorption increased with the dose of ampicillin. An intratracheal dose of 100 mg of probenecid together with 250 mg of ampicillin failed to reduce the absorption of ampicillin from the lungs. The uptake of ampicillin by human foetal bronchial slices in vitro was also studied; although viable, they did not accumulate ampicillin. These preliminary results suggest that ampicillin is absorbed from the lungs by passive diffusion.

Relapse of bacterial infection is a major problem in the chemotherapy of chronic bronchitis (May, 1968). *Haemophilus influenzae*, the important pathogen, is only moderately sensitive to antibacterial drugs, most of which penetrate into bronchial secretions poorly and probably cause the bacteria to be only partially suppressed. An antibiotic often used for the treatment of bronchial infection is ampicillin. Although it is bactericidal against *H. influenzae* in vitro, it is only occasionally able to prevent relapse of *H. influenzae* bronchial infections and even then it has to be given in large doses (May and Delves, 1965). These authors consider this is due to the poor penetration of ampicillin through the blood-bronchial fluid barrier so that bactericidal levels for *H. influenzae* are not reached in respiratory tract secretions. Low levels of ampicillin have also been found in cerebrospinal fluid (Taber, Yow, and Nieberg, 1967) and aqueous humour (Goldman, Broughton, Javed, and Lauderdale, 1973) even when large parenteral doses are administered. These observations are in keeping with the physicochemical properties of ampicillin. It has an even stronger acidic group (pKa=2·52) than benzylpenicillin (pKa=2·72) (Rapson and Bird, 1963) so that at blood pH it would be virtually completely ionized and therefore would be expected to pass through lipoid membrane barriers poorly. Another possible factor which might result in low concentrations of ampicillin in bronchial secretions is an active transport mechanism which pumps the drug out of bronchial fluid. Such a mechanism has been demonstrated for the absorption of organic acids including penicillin from cerebrospinal fluid (Fishman, 1964) and from aqueous humour (Becker, 1960) and is similar to that in the renal proximal tubule.

These experiments were designed to determine whether active reabsorption of ampicillin from the lungs is a factor responsible for the low levels of the drug in sputum.

Evidence is presented in this paper suggesting that ampicillin is absorbed from the lungs by passive diffusion rather than by active transport.

SUBJECT AND METHODS

These studies were performed on a 28-year-old healthy male volunteer (J.L.M.). The subject had fasted overnight and before the intratracheal injection drank a pint of water to ensure that urine could be passed during the experiment.

INTRATRACHEAL INJECTION The subject was seated upright in a chair with the neck extended, and the skin overlying the cricothyroid membrane was infiltrated with 2% lignocaine. After a few minutes 2 ml of 2% lignocaine was injected into the trachea through the cricothyroid membrane. This was followed by vigorous coughing and resulted in numbing of the throat and some loss of voice. A few minutes later a further 2 ml of 2% lignocaine was injected into the trachea to test for abolition of the cough reflex. When

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this had occurred, 50 mg of ampicillin dissolved in 5 ml of sterile pyrogen-free water was injected into the trachea. Venous blood samples were taken after 20 and 40 minutes, 1, 1½, 2, 2½, 3, 3½, and 4 hours and the plasma was separated. All urine passed ½, 1, 1½, and 2 hours after the intratracheal dose was collected and the volume measured.

This procedure was repeated at weekly intervals using doses of 250 mg and 1,250 mg of ampicillin respectively.

The effect of probenecid on ampicillin absorption from the lungs was studied by giving ampicillin 250 mg together with probenecid 100 mg in 5 ml of water by intratracheal injection and taking venous blood samples as before.

BRONCHIAL SLICE TECHNIQUE Cross and Taggart's (1950) renal slice technique for the study of active transport of organic acids by the kidney was modified and used with bronchial slices. This method involved incubating slices of bronchus in Krebs-Ringer solution containing ampicillin and measuring its concentration in the tissue and culture medium after a period of incubation.

Human foetal bronchus was obtained from the Royal Marsden Hospital Organ Culture Bank, London.

Slices of bronchus less than 0·5 mm thick were cut with a razor, dried with blotting paper, and weighed. Bronchial slices weighing approximately 200 mg were divided into four aliquots. Each aliquot was placed on a piece of filter paper (2×3 cm) and floated on 5 ml of culture medium (Krebs-Ringer solution containing 20 μg/ml of ampicillin) in a plastic Petri dish as shown in Figure 1. A stream of gas (95% O_2 and 5% CO_2) was passed into the culture medium beneath the filter paper by piercing the side of the Petri dish with a No. 1 needle. This stream of gas served to keep afloat the filter paper which tended to sink when wet, agitated and buffered the culture medium, and supplied oxygen to the slices. The bronchial slices were incubated at 37°C for 16 hours. After this period they were shown to be viable by the presence of active ciliary movement observed microscopically. Homogenization of the bronchial slices was performed with a glass tissue grinder, and samples of the supernatant together with samples of the culture medium were obtained.

All samples for ampicillin assay were stored at −20°C until analysed. Ampicillin was assayed in the samples, within two days of collection, by the cup-plate method using *Sarcina lutea* as a test organism.

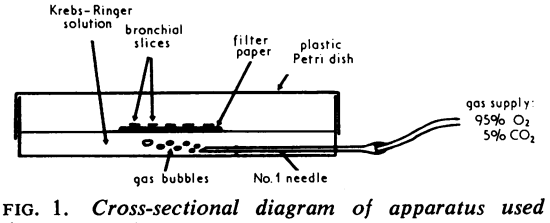

FIG. 1. *Cross-sectional diagram of apparatus used for bronchial slice studies.*

RESULTS

When ampicillin was injected into the trachea it provoked a desire to cough. After doses of 50 mg and 250 mg this was voluntarily suppressed, but after the 1,250 mg dose this was not possible and cough with expectoration occurred; data obtained after this dose have therefore to be interpreted with caution.

Plasma levels of ampicillin obtained after various intratracheal doses are shown in Figure 2. These results show that the plasma levels of ampicillin increase with the intratracheal dose. Another measure of absorption used was the area under the plasma level-time curve. For the three doses the ratio of the areas under the plasma level-time curves was 1 : 3·5 : 23, again showing that absorption of ampicillin increased with the intratracheal dose.

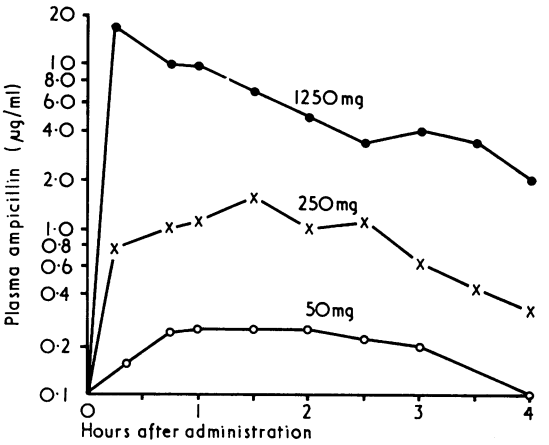

FIG. 2. *Comparison of plasma levels of ampicillin after different intratracheal doses:* ●——● 1,250 *mg;* x——x 250 *mg;* o——o 50 *mg.*

As shown in the Table, the larger the intratracheal dose of ampicillin, the greater is the amount excreted in the urine. Ratios of the cumulative weights of ampicillin excreted in the urine after each of the three doses were as follows: 1 : 7 : 88 (30 minutes); 1 : 5 : 48 (1 hour); 1 : 5 : 42 (1½ hours); and 1: 5 : 35 (2 hours), while the ratio of the doses was 1 : 5 : 25. Considering the urinary excretion of ampicillin after the 50 mg and 250 mg doses only (there was coughing after the 1,250 mg dose), after 1, 1½, and 2 hours the ratio of the

TABLE

CUMULATIVE WEIGHT OF AMPICILLIN (mg) EXCRETED IN URINE AFTER VARIOUS INTRATRACHEAL DOSES

Time after Intratracheal Dose (hr)	Intratracheal Dose (mg)		
	50	250	1,250
0·5	1·2	8·9	105·6
1	4·0	20·9	192·5
1·5	5·8	30·5	242·9
2	7·8	38·9	271·5

cumulative weight of ampicillin excreted in the urine was 1 : 5, which is the ratio of the doses. This is the ratio one would expect to find if ampicillin was absorbed from the lungs by passive diffusion.

Coughing following the 1,250 mg dose of ampicillin would tend to increase its distribution in the lungs and the high rate of absorption after this dose might be due to an increase in the lung surface exposed to this drug.

The effect of probenecid on the plasma levels of ampicillin after intratracheal administration is shown in Figure 3. Plasma levels of ampicillin after an intratracheal injection of ampicillin and

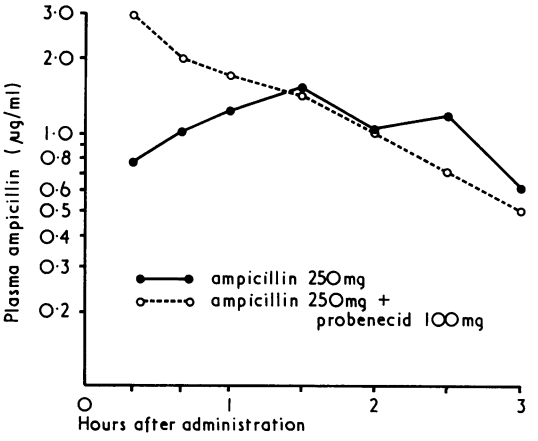

FIG. 3. *Comparison of plasma levels of ampicillin after intratracheal injection of* (a) *ampicillin* 250 *mg* ●——● *and* (b) *ampicillin* 250 *mg* + *probenecid* 100 *mg* o----o.

probenecid were initially higher than after ampicillin alone. The reason for this is not known. Nevertheless these results demonstrate lack of blockade of ampicillin absorption by probenecid.

A typical example of the uptake of ampicillin by human foetal bronchial slices is as follows:

Weight of bronchial slices = 193 mg

Weight of ampicillin in bronchial slices = 1·5 µg

Concentration of ampicillin in bronchial slices $= \frac{1{\cdot}5}{0{\cdot}193} = 7{\cdot}8$ µg/g.

Concentration of ampicillin in medium = 8·0 µg/ml

The bronchial slices were shown to be viable by the presence of active ciliary movement.

These results show no evidence of accumulation of ampicillin by human foetal bronchial slices.

DISCUSSION

Using plasma levels, the area under the plasma level-time curve and urinary excretion as measures of ampicillin absorption from the lungs, the rate of absorption of the drug was found to increase with the intratracheal dose. There is no evidence of rate limitation of absorption as would occur with saturation of an active transport mechanism for organic acids (Schanker, 1962), and probenecid failed to reduce ampicillin absorption from the lungs. These findings, although preliminary, suggest that ampicillin is absorbed from the lungs by passive diffusion. Similar findings were obtained with acidic drugs such as pAH aminohippurate in rats by Enna and Schanker (1970). They found that organic acids were absorbed from the lungs by passive diffusion at rates related to their molecular size and lipid/water partition coefficient.

From the present in vivo studies on pulmonary absorption of ampicillin it is not possible to distinguish between bronchial absorption and alveolar absorption, the results reflecting absorption from both surfaces. The results of the bronchial slice technique, however, provide no evidence of an active transport mechanism for ampicillin in the human bronchus. Similar findings have also been obtained with human bronchus using para-aminohippurate (Maddocks, 1973). These results fail to confirm the suggestion of May and Delves (1965) that ampicillin might be actively secreted by the bronchial mucosa.

Thus it appears that the poor penetration of ampicillin into bronchial secretions is related to its physicochemical properties, particularly its strongly acidic group (pKa 2·52) and its poor solubility in lipids. This gives rise to the question whether the levels of ampicillin in bronchial secretions may be increased by some means. There are two approaches to this problem. First, the permeability of the blood-bronchial fluid barrier might be increased. Buergi, Regli, and Medici (1969)

reported that bromhexine increased the penetration of oxytetracycline into sputum but Ingold and Shaylor (1971) found it had little effect on the penetration of ampicillin into sputum. Secondly, the ampicillin molecule might be modified, provided there is no loss of antibacterial activity, by introducing a suitable functional group to make it a weaker acid (Albert, 1952). Also the ampicillin molecule might be chemically modified, the so-called drug latentiation of Harper (1959), so that the biologically active compound, ampicillin, is released in bronchial secretions.

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