

The effect of methacholine on nasal transmucosal potential difference in normal human subjects*

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SUMMARY

This study was proposed to test the hypothesis that the parasympathetic system might attribute to the transmucosal potential difference. In a double-blind, placebo-controlled study six volunteers had nasal transmucosal potential difference (NTPD) recorded at 4-min intervals during 12-min periods of rest, before and after treatment, as well as during exercise and recovery. Application of placebo did not significantly alter NTPD at rest. There was a significant rise during exercise ($p < 0.05$). The application of methacholine significantly increased NTPD at rest ($p < 0.01$); there was a further rise during exercise ($p < 0.02$). We conclude that parasympathetic stimulation can increase the transmucosal potential difference.

Key words: transmucosal potential difference, methacholine

INTRODUCTION

Transmucosal potential difference is caused by the transmembrane movement of ions, controlled by ion channels. Neurotransmitters are known to regulate the activity of ion channels, possibly through regulation of G-proteins (Dunlap et al., 1987). Nasal mucosal epithelium generates an electrical potential difference, the magnitude of which is known to vary in different regions of the nose as has been demonstrated by Knowles et al. (1981a). In cystic fibrosis the nasal transmucosal potential difference (NTPD) is significantly greater, being negative on the mucosal side (Knowles et al., 1981b). Exercise is also known to increase NTPD (Harris et al., 1990). The purpose of this study is to investigate the influence of cholinergic receptors on NTPD both at rest and during exercise.

MATERIAL AND METHODS

Six female volunteers aged between 21 and 29 years (mean age 23.5) were recruited. They had no history of nasal or respiratory diseases, and were free from respiratory infections for at least 6 weeks. All were non-smokers and were not receiving any medication. NTPD measurements were made using an exploring bridge consisting of a silastic Foley catheter size 8 (Simpla[®]), filled with a 1:1 (v/v)

mixture of Ringer's solution and electrocardiograph electrode cream (Siemens[®]). This resulted in a mixture which ensured contact with the mucosal surface without perfusion of the nasal mucosa, via the aperture at the tip of the catheter. The reference bridge was a Calomel electrode placed over a 3-mm diameter area of lightly abraded skin on the forearm. Readings were made by advancing the tip of the catheter along the floor of the nose, between the inferior turbinate and septum, to the site of maximum stable recorded voltage (PD_{max}), which was usually reached at 3 cm. The catheter was marked at this distance to enable re-insertion to the same place, and readings were taken from this site throughout the study. Recordings from a high-impedance voltage meter were averaged by a computer. Once the site of PD_{max} was established at the beginning of the study, the time taken for each reading – including 5 s for computer averaging – was about 10 s, thus causing minimal interruption of the study. Readings were taken from the right nares in all cases. Subjects in whom there was difficulty placing the catheter due to hypertrophy of the turbinates or septal deflection, or who had difficulty tolerating catheter insertion, causing sneezing or rhinorrhoea, were excluded from the study. NTPD readings were taken at 4-min intervals during

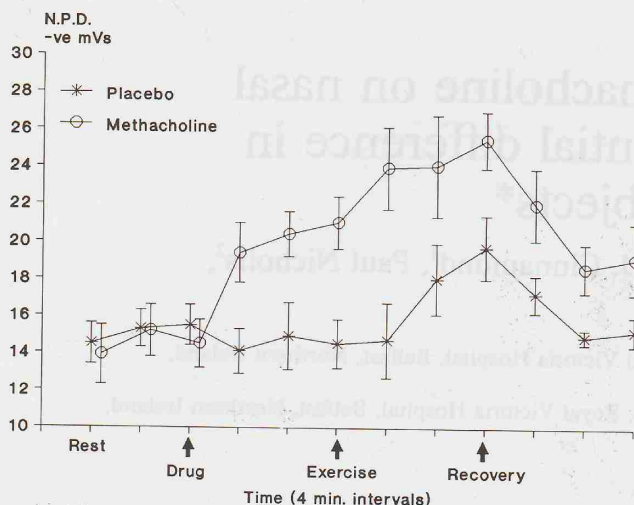


Figure 1. Change in nasal transmucosal potential difference at rest and during exercise, with either placebo or methacholine (error bars indicate SEM).

12 min of rest, before and after treatment, as well as during 12 min of exercise and recovery. The timing of the exercise after application of drug was chosen because preliminary experiments in two subjects demonstrated that the potential difference rise had started to wain by 16 min after drug application.

Each of the six volunteers received in a randomly allocated Latin-square design, double-blind, cross-over study either placebo (0.24 ml of normal saline to each nostril) or methacholine (10 mg in 0.24 ml solution to each nostril). The drugs were supplied by the University Department of Pharmacology in coded containers. The code was broken at the end of the study. The solutions were applied via a pump-action atomizer which delivered 0.12 ml/puff of solution in a mist form. The nozzle was placed past the nasal vestibule and the subject sniffed during delivery of the spray. Exercise was performed on a cycle ergometer, each volunteer achieving a pulse rate of 80% maximum, predicted for age.

The study was approved by the Research Ethical Committee, Faculty of Medicine, The Queens University of Belfast. Statistics were calculated using Student's paired t-test.

RESULTS

The resting values of potential difference on placebo and active drugs were not significantly different ($p > 0.05$; Figure 1). Application of placebo did not alter the potential difference ($p > 0.05$), but methacholine increased it significantly ($p < 0.01$). Exercise significantly increased nasal potential difference in the placebo group ($p < 0.05$). The methacholine group showed a further significant rise ($p < 0.02$) with exercise, and this peak value was significantly greater from the peak value of the placebo group ($p < 0.02$). After exercise both returned to the resting value, neither being significantly different from the pretreatment resting values ($p > 0.05$).

DISCUSSION

These results demonstrate that methacholine increases NTPD at rest in normal subjects. A previous study by Boucher et al. (1980) using canine tracheal mucosa had failed to demonstrate an effect of muscarinic stimulation upon transmucosal potential difference. The agonist used in that experiment was acetylcholine which is rapidly hydrolysed by acetylcholinesterase. The different results reported here may be due to intraspecies differences or to the slower metabolism due to a methyl group instead of hydrogen in the methacholine molecule. Muscarinic receptors have been identified in the glandular acini of human nasal mucosa but not on blood vessels (Van Megen et al., 1991). Physiologically, this is confirmed by the profuse nasal secretion caused by the application of methacholine (Borum, 1979). It is possible that the change in potential difference noted here has been related to the increase in secretion either from the glands or the goblet cells lining the mucosa. It is known that exercise increases nasal secretion production (Stanford and Stanford, 1983), transmucosal potential difference (Harris et al., 1990), and sympathetic activity as indicated by nasal decongestion (Richerson and Seebom, 1967). Muscarinic receptor stimulation is known to regulate transmembrane movement of calcium, potassium and sodium ions, possibly by an action on the G-proteins (North, 1986), but it is not possible in the present experiment to know if the potential difference changes are related to increase of chloride secretion or relatively less absorption. The increased potential difference with exercise is enhanced by prior stimulation with methacholine in the present experiment, suggesting a priming effect, but does not necessarily imply that the exercise effect is itself a straightforward parasympathetic response. Excess secretion from the respiratory tract is a symptom in most respiratory diseases (e.g. rhinitis, chronic bronchitis, asthma, cystic fibrosis). The present observation that parasympathetic stimulation alters the transmucosal potential difference as well as increasing secretion will provide a better understanding of the mucosa and may lead to the development of better therapy.

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SUMMARY

The differing effects of halothane and isoflurane on nasal mucosal blood flow was investigated by means of laser-Doppler flowmetry in a total of fourteen patients who received one of these inhalational agents during anaesthesia. A trend towards lower nasal flow was seen in the halothane group but, due to insufficient numbers significance could not be demonstrated. These changes in flow appeared not to be related to the fall in perfusion pressure which were seen in both groups of patients and were thought to be due to locally vasoactive effects.

Key words: nasal mucosa, blood flow, anaesthesia

INTRODUCTION

During nasal surgery, local vasoconstrictive agents and hypotensive anaesthetics must both be used to decrease capillary blood loss and thereby improve the operating field. The effects of inhalational anaesthetics on nasal mucosal blood flow have not been previously examined, although their effects on flow in many other organs have been extensively studied (Eger, 1981). Both halothane and isoflurane may be used to produce hypotensive anaesthesia, albeit by different actions. This study was designed to detect any significant differences in nasal mucosal blood flow not due to the hypotension produced but due to the specific action of each agent respectively. Our preliminary results are published here.

METHODS

After approval from the Local Ethics Committee, 14 ASA Class I and II patients undergoing elective hypotensive anaesthesia for nasal endoscopy or surgery were selected (Table 1). Patients with a history of hypertension, cardiovascular pathology of any form and patients taking oral anticoagulation or any form of nasal medication were excluded. Written informed consent was obtained prior to participation.

All pre-operative medication with any known effect on patients had anaesthesia induced with 2.5 mg/kg Fentanyl followed by 5 mg/kg Thiopentone, 4 mg/kg Vecuronium was given to facilitate tracheal intubation. Ventilation was

Table 1 Patient data Mean (SD) (range) age, weight, colour index according to group

Group	Halothane (n=7)	Isoflurane (n=7)	P
Age (years)	41 (16.8)	33 (12.1)	0.65
Weight (kg)	72 (18.2)	63 (21.6)	0.75
Mean Colour Index	1.2	1.7	

N.S. = not significant (p > 0.05)

maintained with N₂O:O₂ 50:50 and 100% oxygen using a Stryker Servo-C™ ventilator. The right radial artery cannula was cannulated using an 18-gauge 10-cm arterial catheter-bundle system, catheter being placed in the distal anastomosis as required to maintain the mean arterial value between 5.5 and 6.5 kPa. A radial arterial cannula (18-gauge) was cannulated with a 20-gauge cannula (18-gauge) and arterial central pressure was measured continuously throughout the anaesthesia. The ParvoFlo™ laser-Doppler flowmeter of the device (PeriFlux 5000, Perimed AB, Sweden) was calibrated and placed under the skin surface through a speculum to rest on the nasal mucosa. The mucosa covering the nostril and the nasal septum were exposed. The probe was held in place by a sliding under lip force-repelling arrangement. The distance of the laser beam's range (i.e. distance in arbitrary units of 0 to 1000) The