

THE EFFECT OF VAGOTOMY AND HYDROCORTISONE ADMINISTRATION ON PULMONARY SURFACTANT ACTIVITY IN ADULT ALBINO RATS

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Summary : The role of vagus and hydrocortisone in the regulation of lung surfactant was studied in adult albino rats. Dynamic surface tension and phospholipid content were measured in the lung wash for estimating surfactant activity. The results indicate that vagotomy significantly decreases the surfactant activity whereas hydrocortisone does not alter it. But when hydrocortisone was administered prior to vagotomy it could prevent the decrease in surfactant activity. Thus it is concluded the regulation of lung surfactant in the adult lung is mainly by vagus nerve and hydrocortisone as such has no role but in the absence of vagal regulation hydrocortisone could maintain normal lung surfactant activity.

Key words : vagotomy

hydrocortisone

surfactant activity

INTRODUCTION

Experimental studies in the fetal lamb (4) and rabbit (7) indicate that hydrocortisone accelerates the appearance of lung surfactant. Similarly in a human study (9) a significant decrease in the incidence of Respiratory Distress Syndrome (RDS) was observed in the premature new born when hydrocortisone was administered to their mothers 48 hr prior to their delivery. These observations suggest that hydrocortisone regulates fetal lung surfactant. However in adult lung evidence so far suggests that the regulation of surfactant is by vagus nerve (13). So we conducted a pilot study to observe the effects of hydrocortisone on lung surfactant in adult albino rats by measuring dynamic surface tension of the lung wash and reported that hydrocortisone does not alter the lung surfactant activity (12). Since a single method of assessing the pulmonary surfactant property

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may not give the true picture of the quality and quantity of lung surfactant, the present investigation was planned to study both dynamic surface tension measurement as well as estimation of phospholipid content of lung. In addition it was planned to study the possible inter-relationship between hydrocortisone and vagus in the regulation of lung surfactant

MATERIAL AND METHODS

Healthy albino rats of either sex weighing 140 ± 10 g were divided randomly into four groups of 8 rats each. In group II rats, the effect of hydrocortisone on lung surfactant was observed for 48 hr by injecting hydrocortisone (3 mg/100 g ip.) daily for two consecutive days. In Group III rats the effect of vagotomy on lung surfactant was studied by subjecting the rats to bilateral cervical vagotomy (12) and keeping them in the vagotomised condition for six hours. The effect of hydrocortisone followed by vagotomy was studied in Group IV rats which received hydrocortisone injections as in Group II rats followed by bilateral cervical vagotomy as in Group III rats. Sham operation was performed on Group I rats which served as the controls. At the end of the above experiments, rats of all the groups were sacrificed under nembutal anaesthesia (4 mg/100 g, ip.) and both lungs in TOTO alongwith trachea were dissected out for lung surfactant studies. The lung surfactant was extracted by repeatedly washing the lungs with 0.9% saline through the trachea. The total volume of saline used was 25 ml and saline surfactant solution was then transferred to the trough of a modified Wilhelmy type of surface tension balance (8). The maximum and minimum surface tensions were then measured and the extract stability index (ESI) was calculated using the formula (3).

$$\text{ESI} = \frac{2(\text{T max} - \text{T min})}{(\text{T max} + \text{T min})}$$

Estimation of phospholipids : At the completion of surface tension measurement, the saline surfactant solution in the trough was collected by rinsing the trough with saline. The saline wash was placed in chloroform-methanol mixture (2 : 1, V/V) and lipids were extracted (6). The chloroform extract was evaporated to dryness at 63°C in a stream of nitrogen. The dried extract was digested with 10N H₂SO₄ and phospholipid phosphorous content was determined (14). The phosphorous content in 25 ml was computed for 1 g of wet tissue.

RESULTS

Values of maximum and minimum surface tension of lung surfactant, ESI and phospholipid content in all the groups are shown in Table I. Group II animals subjected to hydrocortisone injection did not show any significant change in surfactant activity or phos-

pholipid content as compared to the control group. Group III animals subjected to bilateral vagotomy showed a significant decrease in phospholipid content and surfactant activity indicated by increased minimum surface tension and decreased ESI as compared to the control group. Group IV animals which received hydrocortisone prior to vagotomy showed a surfactant activity and phospholipid content similar to that of the control group.

TABLE I : Effect of hydrocortisone on surfactant activity of the control and vagotomised rats (MEAN \pm SD, n=8).

| Group/treatment | Dynamic surface tension (dyn/cm) | | Extract stability index | Phospholipids in μ g/g |
|------------------------------|----------------------------------|--------------------|-------------------------|----------------------------|
| | Maximum | Minimum | | |
| I Control | 35.20 \pm 1.30 | 20.78 \pm 1.05 | 0.56 \pm 0.02 | 11.19 \pm 0.38 |
| II Hydrocortisone treated | 34.12 \pm 1.27 | 20.10 \pm 1.20 | 0.54 \pm 0.03 | 11.49 \pm 0.41 |
| III Vagotomy | 35.18 \pm 1.52 | 26.56 \pm 0.82** | 0.35 \pm 0.04** | 9.28 \pm 0.27* |
| IV Hydrocortisone + Vagotomy | 34.78 \pm 1.51 | 19.78 \pm 1.15 | 0.55 \pm 0.05 | 10.54 \pm 0.42 |

** P < 0.001

* P < 0.01

DISCUSSION

Our observation in Group II rats indicate that hydrocortisone does not alter the lung surfactant. This is similar to the observations of Baden *et al.* (1) who have reported that hydrocortisone was ineffective as therapeutic agent in premature new born suffering from R.D.S. But since the earlier reports on experimental animals (4,7) indicate that at least 48 hr are required for hydrocortisone to increase the lung surfactant, Baden *et al.* (1) have suggested that if these premature new borns with R.D.S. would have survived for 48 hours then hydrocortisone could have increased the lung surfactant. In this connection, it is worth mentioning here that effects of increasing the doses of hydrocortisone gradually from 3 mg/100 g to 30 mg/100 g and also increasing the duration of action of hydrocortisone from 48 to 72 hr were studied in the present work in a separate group of rats and and it was observed that the results were similar to group II rats indicating no significant alteration in lung surfactant. In the earlier report of increased fetal lung surfctant following hydrocortisone administration it was suggested that hydrocortisone acts as an enzyme inducer (9). The specific action of hydrocortisone is that it induces phosphorylcholine glyceride transferase leading to increased synthesis of lecithin through the choline incorporation pathway (5) and it is effective only at a particular critical period in the development of fetal lung (9). In lambs (1), it is 130th to 135th day of gestation (full term being 145 days) and in rabbit (7) it is 26th to 27th of gestation (full term being 31 days) and in human fetus (9) just 48 hours before delivery.

From these observations and also from the results of Group II rats of the present study it is suggested that hydrocortisone is effective in increasing lung surfactant only in the fetal lung but not in the new born lung or adult lung.

To rule out the possibilities of degradation of surfactant (half life of surfactant is less than 15 hr) (10) and pulmonary oedema which usually begins to develop two hours following vagotomy (12) as the cause of decreased surfactant in our group III rats, a separate group of rats was studied for surfactant activity and phospholipid content $1\frac{1}{2}$ hr after vagotomy and the results were similar to group III rats. As regarding the regulation of lung surfactant by the vagus, this observation of decreased surfactant in the lung wash following vagotomy and also from the reports (2, 11) of decreased granule content in the type II cells following vagotomy indicates that surfactant decreases both inside the Type II cell also and outside it (in the lung wash). So it is suggested that vagotomy decreases the synthesis of lung surfactant. As mentioned earlier 2 hr after cervical vagotomy extravasation of fluid appears in the alveoli and it gradually increase to such an extent that it kills the animal in about 6-8 hr (12). Since hydrocortisone is known to prevent such extravasation of fluid, a separate group of rats was studied to observe the effects of hydrocortisone on lung surfactant before the development of pulmonary oedema, i.e. $1\frac{1}{2}$ hr after vagotomy and the results showed near normal lung surfactant, as in Gr. IV.

From these observations, it is concluded that the regulation of lung surfactant activity in adult lung is mainly by the vagus nerve and hydrocortisone as such has no role, but in absence of vagal regulation hydrocortisone could maintain the normal lung surfactant activity.

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