

ADENYLATE CYCLASE AND PHOSPHODIESTERASE ACTIVITIES DURING ANAPHYLACTIC SHOCK IN GUINEA PIGS

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Cyclic AMP (adenosine 3,5-monophosphate) is recognized as the intracellular mediator which regulates a number of cellular processes (Robison, Butcher and Sutherland, 1968; Sutherland, Robison and Butcher, 1968). The intracellular level of cyclic AMP is determined by the activity of at least two enzymes. The formation of cyclic AMP from ATP is catalyzed by adenylate cyclase, a particulate enzyme commonly found as a component of the plasma membrane (Sutherland and Rall, 1962).

Recent studies have shown that cyclic AMP plays a major role in the events that activate and regulate immune responses (Ishizuka, Gafni and Braun, 1970; Braun, Ishizuka, Winchurch and Webb, 1972; Gallo and Whang-Peng, 1971; Uzunova and Hanna, 1973).

On the other hand, several lines of investigation have suggested that the cyclic AMP system operates as a second «messenger» in the allergic release of histamine (Bourne, Melmon and Lichtenstein, 1971a; Bourne, Lichtenstein and Melmon, 1971b; Capurro and Levi, 1971; Kaliner, La Raia, Orange, Austen and Austen, 1971; Austen, Orange and Austen, 1971a; Orange, Kaliner, La Raia and Austen, 1971b). Lichtenstein (1971) and others have shown that cyclic AMP inhibits the IgE mediated release of histamine.

These data prompted us to study the adenylate cyclase and phosphodiesterase activities in the lung of guinea pigs during anaphylactic shock. Although many mediators are considered to play a role in the pathogenesis of anaphylaxis, histamine was the first described and remains the most important.

Materials and methods

Randomly bred male guinea pigs were used throughout these experiments. The animals were sensitized by a single intraperitoneal injection of 1 ml of 1% crystalline bovine serum albumin and were challenged two weeks later by i. v. injection of 4 ml of the same antigen solution. The organs were taken out for enzyme determination at the phase of expressed shock symptoms.

Adenylate cyclase activity was measured by the method of Krishna, Weiss and Brodie (1968), which is based on the rate of conversion of ATP-¹⁴C to labelled cyclic AMP. The latter was measured in a liquid scintillation counter after its separation and isolation.

Phosphodiesterase activity was determined by the method of Butcher and Sutherland (1962). In this procedure the 5-AMP formed from cyclic AMP is converted in the presence of excess alkaline phosphatase to adenosine and inorganic pyrophosphate. The latter was measured colorimetrically.

The protein content was determined by the method of Lowry, Rosebrough, Farr and Randall (1951).

Results and conclusions

As can be seen in fig. 1 adelynate cyclase activity in the brain did not change during anaphylactic shock. In contrast to the brain the adelynate

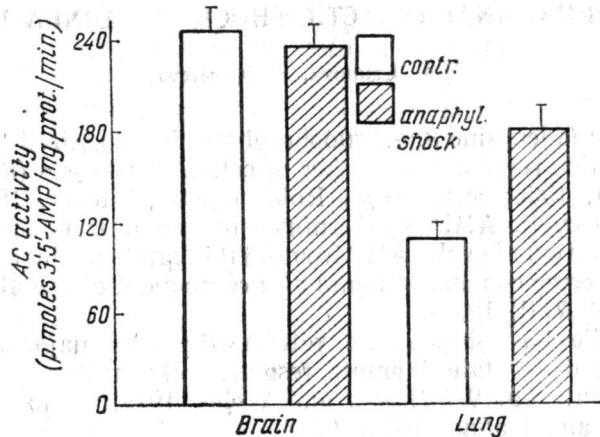


Fig. 1

cyclase activity in the lungs of sensitized guinea pigs increased almost double in comparison with saline treated controls. The difference is statistically significant ($p < 0,01$).

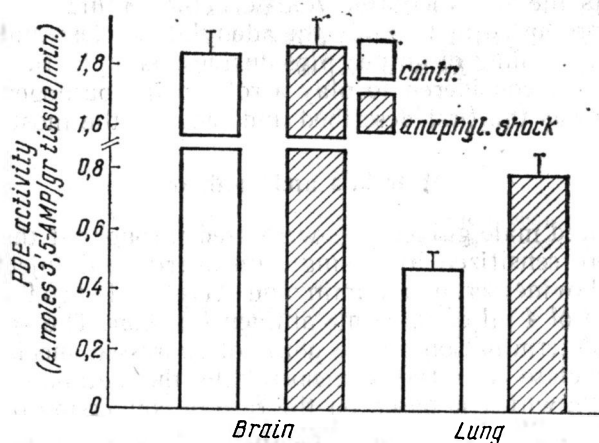


Fig. 2

From fig. 2 it can be seen that there is no difference between the phosphodiesterase activity in the brains of animals with anaphylactic shock and the enzyme activity of the control group; in contrast, phosphodiesterase activity increases by about 30% in the lungs of the sensitized animals which developed

anaphylactic shock. Here again the difference is statistically significant ($p < 0,02$).

Sensitized guinea pigs given i. v. injection of the same antigen used for sensitization undergo an anaphylactic reaction, called anaphylactic shock. The latter is characterized by restlessness, chewing, rubbing of the nose, dyspnoea, convulsive movement and convulsions frequently terminating in death (Kabat and Mayer, 1961). This reaction initiated a series of events leading to damage of the cells of these tissues. By a similar process most cells are altered, resulting in the release of histamine, which plays a major role in the allergic response.

The studies of Lichtenstein (1971), Orange et al. (1971a, b), Kaliner et al. (1971) and others have shown that cyclic AMP inhibits the in vitro release of histamine.

We have demonstrated that both adenylate cyclase and phosphodiesterase activities did not change in brain, whereas the enzyme activity in the lung, which is known to be a target organ in anaphylactic shock, increased markedly.

We suggest that the role of the activated cyclic AMP system in this case is to stop an overreaction of the organism in response to the challenge with the anaphylactogen.

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**АДЕНИЛАТ ЦИКЛАЗНАЯ И ФОСФОДИЭСТЕРАЗНАЯ АКТИВНОСТЬ
У МОРСКИХ СВИНОК В УСЛОВИЯХ АНАФИЛАКТИЧЕСКОГО ШОКА***А. Узунова, З. Кемилева***Р Е З Ю М Е**

Исследована активность аденилат циклазы и фосфодиэстеразы в легком и в мозгу морских свинок, находящихся в состоянии анафилактического шока. Устанавливается повышенная активность обоих энзимов в легком, но не и в мозгу. Авторы полагают, что повышенная энзимная активность является защитным механизмом, вызывающим образование циклического АМФ; последний, со своей стороны, действует как ингибитор освобождения гистамина.