EFFECT OF NON-NARCOTIC ANALGESIC DRUG (ASPIRIN) ON DEVELOPING CHICK EMBRYO

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ABSTRACT

The administration of low dosage of aspirin in early developing embryos of chick demonstrated severe overall stunted growth and abnormalities in certain organs as compared with the embryos received at later stages of development. Aspirin administration also resulted in drastic changes in the shell calcium, permeability rate of shell, carbohydrates, proteins, amino acids and lactate of developing embryo.

INTRODUCTION

ASPIRIN-LIKE analgesics are used for relief of the mild to moderate headache, muscle ache, arthralgia etc acting primarily at the site of origin of pain. They may be antipyretic and/or antiinflammatory. Aspirin (acetyl salicylic acid) is an effective analgesic and antipyretic, and is useful for the treatment of rheumatic diseases although it has no effect on associated cardiac and visceral complications. It is consumed in larger quantity than any other therapeutic agent. The wide availability and occasionally indiscriminate use of this drug has led to an underestimation of its danger. It accounted for about 4.4% of all drug poisoning and about 7.5% childhood poisoning in 1974. Chronic use of high doses may have a number of gastrointestinal effects. Embryo toxicity and teratogenicity in the pregnant mammals were studied by the administration of the aspirin[1–5].

Recently reports have appeared regarding the influence of aspirin on the reproductive performance and growth of the rat embryos[6,7]. The effect of certain chemicals on the developing chick embryos has been tested[8–15], but work on the development of chick embryo and its biochemical aspects with special reference to the aspirin is lacking. Chick embryos can be used as an experimental model to study the effects of certain drugs on the foetus among the pregnant women. In the present paper, we have studied the effect of aspirin on the developing chick embryos.

MATERIALS AND METHODS

Fertile leghorn eggs were obtained locally from the veterinary college and incubated for 5 days. They were then divided into four batches of 10 eggs each. Aspirin tablets were dissolved in distilled water (100 mg of aspirin in 10 ml of distilled water). The eggs were cleaned with 70% alcohol. A hole was made with a hypodermic needle (sterilized) on the broad end of the eggs. The first batch (control) of the eggs was injected with 0.05 ml of distilled water into the yolk sac at zero of incubation and sealed with paraffin wax. Out of the remaining three batches of eggs (experimental), one batch was injected with 0.05 ml of aspirin per egg (0.5 mg) after 15 days of development, and incubated for further development. Similarly, the other two batches were injected with the same concentration of aspirin (0.05 ml) after 10 and 5 days of development respectively and left for further development. Candling was done daily to observe the egg development. The eggs were rotated twice a day (60% humidity) and the temperature was maintained at 37° ± 1°C

The embryos were collected at the end of the development (after the 20th day). On opening the egg, the embryo was visible in the amniotic sac. The allontoic stalk was cut and the embryos were transferred to a Petri dish, containing 0.85% saline solution. These embryos were checked for the abnormalities by the method of Hamberger and Hamilton[16]. The embryos were weighed and the different biochemical parameters like shell calcium[17], proteins[18], carbohydrates[19], amino-acids[20], lactate[21] were determined. The shell permeability rate was determined with glucose mobilization from shell (containing hypertonic glucose solution) to the external medium (hypotonic solution) and the glucose content was estimated[19]. The results were statistically analysed using the student's *t* test.

RESULTS AND DISCUSSION

Aspirin induced pronounced morphological and biochemical changes during the development of chicks (figure 1a, table 1). The development of

Table 1 *Changes in embryo weight, shell calcium, permeability rate, carbohydrates, proteins, amino acids and lactate in control and aspirin treated chick embryos*

Parameters	Batch I (control)	Batch II	Batch III	Batch IV	Unincubated
Embryo weight (g)	22.18 ± 1.33	13.50* ± 0.69 − 39.13%	6 60* ± 0.37 − 70.24%	5.37* ± 0.27 − 75.78%	–
Shell calcium (mg/g wt of shell)	32.41 ± 1.51	41.13* ± 1.03 27.27%	48.16* ± 0.52 48.50%	50.17* ± 0.36 54.79%	52.86 ± 1.67
Permeability (mg/g of glucose/hr)	66.46 ± 2.62	58.05* ± 0.51 − 11.76%	45.99* ± 2.75 −30.85%	37.89* ± 0.62 −41.85%	35.57 ± 2.21
Carbohydrates (mg/g wt of tissue)	3.11 ± 0.25	2.61* ± 0.18 −27.23%	2.53* ± 0.17 −18.48%	2.26* ± 0.15 −19.42%	–
Proteins (mg/g wt of tissue)	34.23 ± 2.43	27.26* ± 2.96 −20.33%	23.01* ± 2.15 −32.77%	18.29* ± 0.38 −46.56%	–
Amino acids (mg/g wt of tissue)	3.80 ± 0.19	4.30* ± 0.31 13.61%	6.44* ± 0.32 69.47%	7.37* ± 0.28 88.94%	–
Lactate (mg/g wt of tissue)	3.30 ± 2.72	3.61** ± 0.19 9.40%	4.16** ± 0.11 26.06%	4.92** ± 0.12 48.78%	–

The values are mean ± S.D for six individual observations; + and − indicate per cent increase and decrease over control; *$P<0.001$; **Not significant.

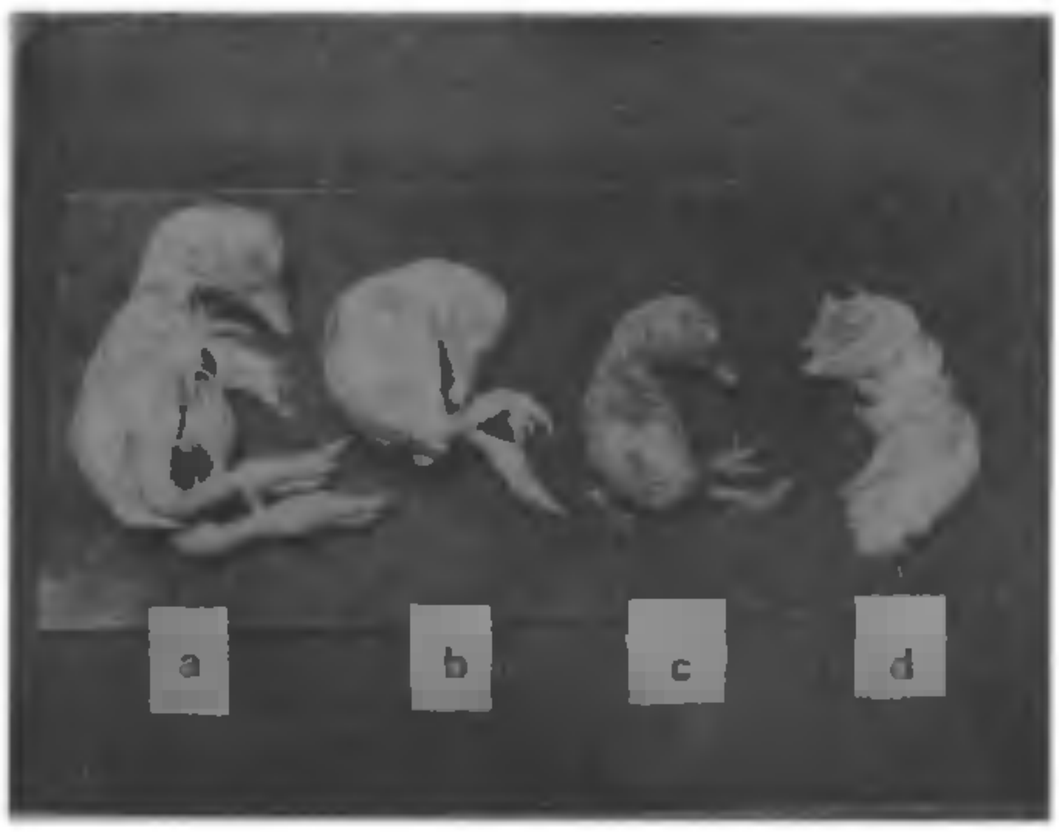

Figure 1a–d. a. Embryo from control egg (note the full development). Embryo administered with 0.5 mg of aspirin after; **b.** 15 days; **c.** 10 days and **d.** 5 days of development.

embryo is reduced to 75% in batch IV, 70% in batch III which received aspirin after 5 and 10 days of development respectively. The reduction in the weight of batch II embryos of 39% is a clear indication that aspirin exerted its effect on growth. The retardation of embryonic growth may be due to its effect on general metabolism. However, experimental embryos which received aspirin at different intervals continued to develop with number of abnormalities. These changes include an overall stunted growth, reduced head circumference, poor development of feathers (figure 1b); shortening of limbs, poor development of head, feathers (figure 1c); poor development of eyes (micropthalalmia), non-development of fingers, degeneration of feathers and poor development of beak (hypognatha) (figure 1d).

The growth retardation in experimental embryos is further supported by the higher amounts of shell calcium, suggesting that the embryos did not utilize the shell calcium for their organogenesis. The experimental embryos' shells have shown lower levels of permeability, when compared with those of control. This directly supports the increase in shell calcium. The developing embryo needs more oxygen as it grows and this is obtained by the thinning and increased porous nature of the shells. As the experimental embryos ceased to grow, the non-

utilization of shell calcium might have altered the oxidative metabolism greatly. Thus the developing embryo may be shifting towards anaerobiosis due to accumulation of lactate content with aspirin injection. Since any stress results the utilization of carbohydrates[22,23], the significant decrease ($P<0.001$) in the levels of carbohydrates was noticed in aspirin injected embryos.

The significant decrease ($P<0.001$) in protein content and increase ($P<0.001$) in amino acid content may be ascribed with the increased activity potentials of proteases in experimental embryos. Several workers have indicated that aspirin exerts its effect acting chiefly on the carbohydrate, protein and lipid metabolism in animals[24–27]. The present investigation suggests that aspirin administration at an early stage of development can cause greater deleterious effects on the growth and metabolism than administration at later stages.

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