EFFECT OF ACTH AND SODIUM SALICYLATE ON THE URINARY URIC ACID : CREATININE RATIO, AND CIRCULATING EOSINOPHILS IN MAN

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(RECEIVED FOR PUBLICATION OCTOBER 6, 1953)

Since Cochran and others (1950) reported the development of Cushing's syndrome in a 12-year-old girl receiving 5 g. aspirin daily for acute rheumatism, much work has focused on the possibility that the therapeutic and metabolic effects accompanying salicylate administration may be dependent upon the intermediary production of ACTH and/or adrenal corticoids. These authors noted frequent past references to the development of acne and puffiness of the face in patients receiving salicylates for acute rheumatism, side-effects commonly seen during ACTH or cortisone therapy. Referring to their single case, together with the previous work of Reid, Watson, and Sproull (1950), they noted similarities in the effects of salicylates and cortisone not only in the therapeutic response in acute rheumatism, but in their metabolic effects on fluid, chloride, nitrogen balance, and plasma proteins.

Many other reports have since been published on the comparative effects of salicylates and ACTH or cortisone in experimental animals and man. These results may be reviewed according as the changes induced by these two groups of drugs are (a) similar, (b) dissimilar, (c) conflicting.

(a) Similar Changes

(i) *Adrenal Ascorbic Acid and Cholesterol.*—Administration of ACTH to rats has been found to decrease the adrenal cholesterol (Sayers and others, 1944a) and ascorbic acid (Sayers and others, 1944b), and the adrenal content of these substances has since been used as a measure of ACTH and cortical activity. Pre-treatment with adrenal cortical hormones protects against this ACTH→depletion effect (Sayers and Sayers, 1947).

Blanchard and others (1950) showed that salicylate caused a significant fall in adrenal ascorbic acid in rats; Hetzel and Hine (1951) confirmed this and showed that the effect was proportional to the dose and that it was inhibited by pre-treatment with cortisone. Van Cauwenberge and Heusghem (1951a) reported depletion of adrenal cholesterol as well as ascorbic acid, and Robinson

(1951) showed that this cholesterol depletion occurred within 30 minutes of salicylate administration.

(ii) *Urinary Uric Acid Excretion.*—It has been known for many years that salicylates in large dosage can increase urinary uric acid excretion (Byasson, 1877; Sée, 1877; Salomé, 1885), and sodium salicylate has been used to maintain the serum uric acid in gouty patients at normal levels for long periods (Marson, 1953). More recently, ACTH and cortisone have been shown to have uricosuric effects (Thorn and others, 1947b), and the increase in urinary uric acid : creatinine ratio after ACTH injection has been incorporated in a test for adrenocortical function (Thorn and others, 1948). Certain authors, believing that the uricosuric effect of salicylate is mediated *via* the pituitary and suprarenals, have described an intravenous sodium salicylate test for investigation of the function of the two glands (Roskam and others, 1951). The test resembles the ACTH test with the replacement of this drug by sodium salicylate. Creatinine excretion is unaffected by ACTH therapy (Mason and others, 1948), and is stated to be slightly increased by salicylates (Hanzlik, 1927).

(iii) *Anti-hyaluronidase Effect.*—Cortisone inhibits the spreading phenomenon (Opsahl, 1949) and both ACTH and cortisone have been shown to inhibit the action of hyaluronidase in causing increased capillary permeability (Benditt and others, 1950). Salicylate has also a definite antidiffusive action (Guerra, 1946; Bertolani and Bergamini, 1950; Schuman and Finestone, 1950; Pelloja, 1952). It does not inhibit hyaluronidase *in vitro* (Swyer, 1948).

(iv) *Arthus Phenomenon.*—Salicylates decrease the Arthus phenomenon (Fischel, 1947), as do also ACTH and cortisone (Germuth and others, 1951). All three drugs inhibit tissue reactivity to bacterial antigen in rabbits (Shwartzman and others, 1950).

(v) *Serum Arteritis.*—Experimental serum arteritis may be inhibited by both cortisone (Rich and others, 1950; Seifter and others, 1950) and salicylates (Macgregor and Wood, 1950; Sullivan and others, 1948).

(vi) *Effects of Hypophysectomy.*—To investigate whether the possible effect of salicylate in stimulating the adrenal corticoids was direct or mediated *via* the anterior

pituitary, various workers have studied the metabolic and haematological effects of salicylate in hypophysectomized animals. Hetzel and Hine (1951) found that salicylate failed to deplete adrenal ascorbic acid in hypophysectomized rats. Van Cauwenberge (1951) confirmed this fact and found that adrenal cholesterol was similarly unaffected. Pelloja (1952) reported that salicylates failed to inhibit the spreading factor in hypophysectomized or adrenalectomized rats.

(b) Dissimilar Changes

(i) *Carbohydrate Metabolism*.—Ingle (1941) showed that cortisone could induce severe glycosuria and hyperglycaemia in normal rats fed on a high carbohydrate diet, and ACTH was later found to have similar effects (Ingle and others, 1946). ACTH raises the fasting blood sugar levels in man (Forsham and others, 1948), and the similar effect with cortisone is now utilized in the protection against hypoglycaemia in Addison's disease. Many reports in the last quarter of the 19th century stated that salicylates in large dosage decreased diabetic glycosuria, and for a time these drugs were used in the treatment of this condition (Gross and Greenberg, 1948). Recent reports have shown that salicylates in large dosage reduce the glycosuria of diabetic rats (Ingle, 1950), and lower the blood glucose level (Smith and others, 1952). Smith (1952) showed that the simultaneous administration of salicylates reduced the glycosuric and hyperglycaemic effects of cortisone administered to normal rats fed on a high carbohydrate diet.

Cortisone produces a significant deposition of liver glycogen in rats (Smith, 1952). Salicylate has the reverse effect (Lutwak-Mann, 1942), and also prevents the formation of new liver glycogen by cortisone (Smith, 1952).

(ii) *Adrenal Steroids*.—ACTH administration increases the urinary excretion of both 17-ketosteroids and 11-oxy-steroids (Mason and others, 1947; Thorn and others, 1947a). Bertolani and others (1951) reported that salicylates increased the urinary output of 17-ketosteroids in guinea-pigs. Van Cauwenberge and Heusghem (1951b) found that aspirin caused an increased urinary excretion of reducing steroids in man, but no constant change in the 17-ketosteroid excretion.

(c) Conflicting Reports

Eosinophil Depression.—Hills and others (1948) first reported a fall in circulating eosinophils after an injection of ACTH and this response has since been used extensively as an index of adrenocortical function (Thorn and others, 1948, 1951). Kelemen and others (1950) reported that single doses of 6-10 g. salicylate induced a significant fall in circulating eosinophils in man, and Bertolani and others (1951) noted a similar eosinopenic effect in guinea-pigs. Meade and Smith (1951) failed to demonstrate any significant change in the eosinophil counts in normal persons within 4 hrs of administration of 75 gr. sodium salicylate. It is noted that this dosage is less than that administered by Kelemen and others (1950).

Roskam and others (1951), however, found that 4-6 g.

sodium salicylate did produce an eosinophil depression, but not until between 4-6 hrs. They suggested that the delayed effect resulted from slow intestinal absorption, as 4 g. sodium salicylate induced eosinophil changes within 4 hrs when given intravenously. It is interesting that these authors had shown that the increased urinary uric acid : creatinine ratio reached its peak within the first 2 hrs of giving salicylate orally. Despite this marked difference in time of occurrence, the authors considered that both the eosinophil and uric acid changes indicated stimulation of the suprarenal cortex.

There are, therefore, many similarities in the metabolic effects produced by salicylate in heavy dosage and those produced by ACTH or cortisone, and also a few dissimilarities, notably those affecting carbohydrate metabolism and urinary 17-ketosteroid excretion. These dissimilarities are probably sufficient to negative the hypothesis that the actions of salicylates are dependent upon intermediary production of ACTH. This is supported by the fact that, with the exceptions of acute rheumatism and chronic gout, salicylates compare unfavourably with ACTH and cortisone in the field of therapeutics.

Present Investigations

The following work fails to confirm the occurrence of eosinopenia during salicylate therapy, and suggests that the increased urinary uric acid : creatinine ratio following salicylate administration is unlikely to result from intermediary ACTH production.

Material.—Experiments were carried out on three female subjects, aged 49, 52, and 53 years, who were suffering from osteo-arthritis, rheumatoid arthritis, and gout respectively, and on three male subjects, aged 31, 31 and 32 years, one with rheumatoid arthritis and the other two with gout.

Method.—All subjects were in hospital receiving normal mixed diets together with 3 pints fluid per 24 hrs. No drugs were prescribed before the tests. On test days the subjects remained in bed and their diet and fluid intake was limited to 10 oz. milk at 6, 10 a.m., noon, 2, and 4 p.m. On these days the bladder was emptied at 6 a.m. and thereafter 2-hourly until 4 p.m. The test days were as follows:

- (a) *Control*.—10 oz. water at 8 a.m.
- (b) *Sodium Salicylate*.—100 gr. freshly prepared, administered orally in 10 oz. water at 8 a.m.
- (c) *ACTH*.—50 mg. given intramuscularly at 8 a.m., together with 10 oz. water by mouth.

On test days (b) and (c), which were separated by at least 3 clear days, the circulating eosinophils were counted just before the administration of the dose and at 9, 10 a.m., noon, 2, 4, and 5 p.m. The counts were made on samples of venous blood by a modification of Randolph's method using *Fuchs-Rosenthal* counting-chambers.

Fig. 1.—Mean values of eosinophil counts in six subjects after administration of:

- (a) 100 gr. sodium salicylate orally,
(b) 50 mg. ACTH intramuscularly.

Fig. 2.—Mean values of urinary uric acid:creatinine ratios in six subjects:

- (a) during control period,
(b) after 100 gr. sodium salicylate orally,
(c) after 50 mg. ACTH intramuscularly.
Results charted at middle of 2-hour periods.

Fig. 1 shows the results expressed as a percentage of the initial values.

The 2-hr urine samples were tested for uric acid (Brown's colorimetric method, 1945) and creatinine (Folin's method, 1914), and the values have been calculated as uric acid:creatinine ratios. Fig. 2 shows the results expressed as percentages of the initial values.

Additional Experiments.—The above tests, with the omission of eosinophil counts, were each performed twice on a female patient aged 48 years, with severe Simmonds' disease.

She was fit until the birth of her fourth child 13 years previously. Lactation then failed completely and since that time she had noticed complete amenorrhoea, extreme fatigue, mental sluggishness, pallor, and loss of axillary and pubic hair. She had suffered two bouts of unconsciousness and in one of these the rectal temperature had fallen to 86° F. Her appearance was typical of Simmonds' disease, the blood pressure was 100-70 mm. Hg, and the laboratory findings included a urinary 17-ketosteroid excretion of less than 1 mg./24 hrs, a basal metabolic rate of minus 39 per cent., and an insulin tolerance test showing marked hypoglycaemic unresponsiveness.

Fig. 3 shows the effect of sodium salicylate and ACTH

Fig. 3.—Mean values of urinary uric acid:creatinine ratios in a case of Simmonds' disease: (a) during control period, (b) after 100 gr. sodium salicylate orally, (c) after 50 mg. ACTH intramuscularly.

Results, charted at middle of 2-hour periods, represent mean of two values.

administration upon the uric acid : creatinine ratios in this patient.

In a further test, three patients (one male aged 31 and two females aged 53), suffering from rheumatoid arthritis, had daily circulating eosinophil counts performed at 10 a.m. and 4 p.m. during a period of 14-15 days. They remained in bed during the test and received a normal mixed diet. No drugs were administered apart from freshly prepared sodium salicylate which was given for a 7-day period in a dosage of 30 gr. at 6 a.m., 2, and 10 p.m. (Fig. 4).

Fig. 4.—Daily circulating eosinophil levels in three subjects expressed as mean of counts performed at 10 a.m. and 4 p.m. During a 7-day period sodium salicylate was administered 30 gr. 8-hrly.

Results

Fig. 1 shows that the ACTH injection (50 mg. intramuscularly) was followed by a depression of the eosinophil level after the first hour, which fell to less than 50 per cent. of the initial value at the 4th and 6th hours. Sodium salicylate administration (100 gr. orally) was followed by an insignificant decrease in the eosinophils at the 4th hour (1.25 per cent. of initial value) and all other counts showed a rise above the initial value.

Fig. 2 shows that ACTH administration was followed by an increase of the urinary uric acid : creatinine ratio, which reached a maximum of 65 per cent. between the 4th and 6th hours, whereas

sodium salicylate administration was followed by a maximum increase of 256 per cent. occurring within the first 2 hours.

Fig. 3 shows that while ACTH (50 mg. intramuscularly) was followed by a slight increase in urinary uric acid : creatinine ratio (maximum 15 per cent. at 6-8 hours) in the patient with Simmonds' disease, sodium salicylate (100 gr. orally) was followed by a considerable increase in the ratio (maximum 147 per cent. at 2-4 hours).

Fig. 4 shows that daily eosinophil counts revealed no significant depression in three subjects during a 7-day period in which sodium salicylate was administered in a dose of 30 gr. 8-hourly.

Discussion

The results show that sodium salicylate had a much greater effect on increasing the urinary uric acid-creatinine ratio than ACTH, and acted more quickly. ACTH had the usual effect in depressing the eosinophil count, whereas salicylate failed to depress eosinophil levels during the 9 hours after administration.

If the uricosuric effect of salicylate is dependent upon ACTH production, one would have to postulate its acting far more quickly than 50 mg. intramuscularly ACTH, and producing ACTH far in excess of the equivalent of the 50 mg. given intramuscularly. This being so, it would be difficult to explain why the eosinophils were not more reduced than with the ACTH, more quickly—and certainly within 4 hours, whereas in fact, no depression was observed within 9 hours of salicylate administration. This observation was reinforced by the failure of sodium salicylate (90 gr. daily) to induce a significant eosinophil depression during a period of 7 days.

In the patient with severe Simmonds' disease, one would expect a greatly diminished response to salicylate if its action were mediated via ACTH production. The uricosuric response, however, was well marked, and greatly exceeded that produced by 50 mg. ACTH intramuscularly.

These results indicate that the uricosuric effects of salicylates in man cannot be accounted for by intermediary production of ACTH, and cannot, therefore, be used in the investigation of the hypothalamus—pituitary—suprarenal system as suggested by Roskam and others (1951).

Summary

The literature dealing with the comparative metabolic and haematological effects of ACTH and salicylates is reviewed.

It is shown that the uricosuric action of salicylates in man cannot be accounted for by the intermediary

production of ACTH, and may not therefore be utilized in the assessment of pituitary-adrenal function.

Sodium salicylate failed to decrease the circulating eosinophils either within 9 hours of the administration of a single dose of 100 gr., or during a 7-day period in which 90 gr. were given daily.

I am indebted to Professor A. P. Thomson in whose Department this work was carried out, to Dr. R. Gaddie and Miss Jean Morris for the biochemical data, and to Dr. M. J. Meynell, clinical pathologist, for the eosinophil counts.

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Effet de l'ACTH et du salicylate de soude sur l'acide urique urinaire, le taux de créatine et les éosinophiles sanguins chez l'homme

RÉSUMÉ

On passe en revue la littérature sur les effets métaboliques et hématologiques comparés de l'ACTH et des salicylates.

On montre que l'action uricosurique des salicylates chez l'homme ne peut pas s'expliquer par la production intermédiaire d'ACTH et on ne peut donc pas s'en servir pour déterminer la fonction surrénopituitaire.

Le salicylate de soude n'a pas réussi à faire baisser les éosinophiles sanguins au cours des neuf heures après l'administration d'une dose unique de 6,5 grammes ni au cours de l'administration quotidienne de 5,8 grammes pendant sept jours.

Efecto de la ACTH y del salicilato de sodio sobre el ácido úrico urinario, la tasa de la creatina y los eosinófilos sanguíneos en el hombre

SUMARIO

Se pasa en revista la literatura sobre los efectos metabólicos y hematológicos comparados de la ACTH y de los salicilatos.

Se demuestra que la acción uricosúrica de los salicilatos en el hombre no se puede explicar por la producción intermedia de la ACTH y que no se puede, pues utilizar para determinar la función suprarrenal-pituitaria.

Con el salicilato de sodio no se consiguió la caída de los eosinófilos sanguíneos dentro de las nueve horas que siguieron la administración de la dosis única de 6,5 gramos ni durante los siete días de administración diaria de 5,8 gramos.