sorption and neotetrazolium reduction. That this mineralocorticoid possesses ultraviolet absorption and the one described by Tait, et al.,[2] did not is the only known differentiating feature between these strikingly similar compounds. A comparison of the two mineralocorticoids is shown in Table II.

A detailed description of this work, including the chromatographic characterization of the mineralocorticoid in numerous solvent systems developed in our laboratory and infrared data on the mineralocorticoid and its polyacetate, will be published soon.

The authors wish to thank Mr. Norbert E. Mayer for technical assistance.

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　　　　RECEIVED AUGUST 20, 1953

ELECTRODES WITH CONVECTION CONTROLLED LIMITING CURRENTS ("CONVECTION ELECTRODES")

Sir:

Experimental conditions were found under which convection controlled limiting currents at the rotated platinum wire electrode are observed. In addition, a "convection" mercury microelectrode has been developed. In stirred solutions the thickness of the boundary layer at an electrode decreases with increasing rates of stirring and eventually reaches a limiting value of "molecular"dimensions. Under these conditions limiting currents no longer depend on the diffusion coefficient of the electroactive species but on the rate of convective transport to the electrode surface. Assuming that τ microliter of solution is transported per second to a unit area of the electrode, the convection controlled limiting current (corrected for residual currents and expressed in microamperes) is postulated to be:

$$i_{conv.} = nFa\tau C = knC \qquad (1)$$

where nF represents the number of coulombs per mole involved in the electrode reaction, C is the bulk concentration of the electroactive species in moles per liter and A the area of the electrode expressed in sq. cm. $-\tau$ is called the "convection coefficient," expressed in cm. $\times$ sec.$^{-1}$, and depends on the prevailing specific hydrodynamic conditions.

A platinum wire electrode, 0.5 mm. in diameter and 5 mm. in length, rotated at 600 to 900 r.p.m., yielded convection controlled limiting currents when the solution was stirred with the aid of a rectangular propeller with four blades (13 $\times$ 5 $\times$ 1 mm. each), mounted concentrically 2 cm. above the electrode and rotated at the same rate as the latter. Identical convection controlled limiting currents were obtained in equinormal solutions of iodide, triiodide and an ethylenediamine tetraacetate complex of thallium(III), the diffusion coefficient of the latter being about one-twentieth as large as that of iodide.

Well-defined limiting currents were obtained with a stationary mercury microelectrode which is being further studied and which consisted of 0.005 ml. of mercury in a 2-mm. bore capillary cuvette attached to the bottom of a 250-ml. electrolysis

cell, the solution being stirred at 900–1200 r.p.m. In 0.1 M potassium chloride as supporting electrolyte the convection current of thallium(I) was proportional to concentration in the range between 5 $\times$ 10^{-6} and 5 $\times$ 10^{-3} M and about twenty times as large as the corresponding diffusion current at the dropping mercury electrode. The half wave potential was independent of concentration at a given rate of change of applied voltage.

The value of the convection coefficient, τ, was estimated from convection currents at 900 r.p.m. for the rotated platinum wire electrode and the stationary mercury microelectrode as 19 and 18 cm. $\times$ sec.$^{-1}$, respectively.

Most prominent advantages of convection electrodes are: (1) stationary electrodes can be used. (2) The limiting current is independent of the diffusion coefficient of the electroactive species. (3) The use of a synchronous motor is not necessary.

Acknowledgment is made to the Graduate School of the University of Minnesota for a grant in partial support of this work.

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PROPERTIES OF A SODIUM-RETAINING PRINCIPLE FROM BEEF ADRENAL EXTRACT

Sir:

Current interest in the amorphous fraction of adrenal extracts[1] makes it desirable to present results of a study of this fraction since further work must await processing of more material.

The extract (furnished by the Upjohn Co.) of 1200 lb. of beef glands was chromatographed on a column of silica gel with the propylene glycol–toluene system. The active fractions, assayed in rats using a method similar to that of Simpson and Tait[2] were combined and rechromatographed. The active fraction was acetylated and chromatographed successively in the formamide–benzene and formamide–benzene–cyclohexane systems. It was then chromatographed three times in the latter system on paper.

The acetate isolated weighed 13.2 mg. and assay for α-ketol with blue tetrazolium indicated 13.2 mg. in terms of cortisone. It could not be crystallized. Its biological activity in dog and rat assays was similar in type and approximately equal to that of desoxycorticosterone acetate. Absorption maxima at 239 mμ ($E_{1\,cm.}^{1\%} = 370$, methanol) and at 1618 and 1671 cm.$^{-1}$ (chloroform) indicated the presence of a Δ^4-3-keto group assuming the substance is a steroid. An acetylated α-ketol side chain was evidenced by absorption maxima at 1736 and 1750 cm.$^{-1}$ (chloroform). The absence of absorption maxima near 1710 (CS$_2$) and 3600 cm.$^{-1}$ denoted absence of an 11-keto and of unacetylated hydroxyl groups. A negative Porter–Silber[3] reaction indicated absence of a 17-hydroxyl group, of a 16–17 double bond, and probably of a 16-acetoxy group.

(1) H. M. Grundy, S. A. Simpson, J. F. Tait and M. Woodford, *Acta Endocrinologia*, **11**, 199 (1952).

(2) S. A. Simpson and J. F. Tait, *Endocrinology*, **50**, 150 (1952).

(3) C. C. Porter and R. H. Silber, *J. Biol. Chem.*, **185**, 201 (1950).

The latter would be expected to give 16–17 unsaturation readily; 3α,21-diacetoxy-Δ^{16}-pregnene-11, 20-dione gives a strong Porter–Silber reaction. The sulfuric acid spectrum showed an absorption maximum at 287 (Δ^4-3-keto group) shoulder at 390 mμ.

The mobility of the free substance and its acetate suggests the presence of four, possibly five, oxygen atoms, two of which are in acetylatable hydroxyl groups. In the propylene glycol–cyclohexane system the acetate moved on paper at approximately the same rate as 11-dehydrocorticosterone acetate and considerably faster than substance S acetate. When the 3-(2,4-dinitrophenylhydrazone) of the acetate was heated one hour at 60° in 0.02 N perchloric acid in acetic acid the absorption maximum at 385 mμ did not change in position. This result demonstrated the absence of a 6-acetoxy group since in parallel experiments with 6α- and 6β-acetoxy-11-desoxycorticosterone 21-acetate 3-(2,4-dinitrophenylhydrazones) the absorption maxima shifted from 387 and 381 to 398 and 400 mμ. These conditions would be expected to cause loss of acetic acid from positions 1, 2 or 7 with increase in the wave length of the absorption maximum.

Hydrolysis of the acetate with citrus acetylase (courtesy of B. C. Bocklage and E. A. Doisy) gave a product which was approximately 85 times as active in the bioassay as desoxycorticosterone acetate. Paper chromatography showed the presence of two substances, one apparently completely deacetylated; crystals (m.p. 163–164°, λ$_{max}^{MeOH}$ 240 mμ) of approximately the same activity as the crude mixture were obtained from the dry ether. The second substance, which could be converted to the first by further treatment with acetylase, crystallized from acetone–petroleum ether, m.p. 217–219°, λ$_{max.}^{MeOH}$ 239 mμ, and was approximately 25 times as active as desoxycorticosterone acetate.

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STRUCTURE OF LIGANDS IN INORGANIC COÖRDINATION COMPOUNDS BY INFRARED SPECTRA
Sir:

In addition to the active frequencies we have observed the forbidden frequencies as weak infrared absorption for such symmetrical ions as NO_3^-, SO_4^{--}, etc., when these ions are present outside the coördination sphere in complex compounds. For example, the totally symmetric frequency of the nitrate ion at 1050 cm.$^{-1}$ has been observed as a weak infrared absorption in the complex, $[Cu\{SC(NHCH_2)_2\}_4]NO_3$, and that of the sulfate ion at 980 cm.$^{-1}$ in the compound $[Cu(NH_3)_4]\cdot SO_4\cdot H_2O$. The appearance of these forbidden symmetrical frequencies in absorption can be explained as due to the deformation of the ions in the molecular field of the crystals.

If such ions are coördinated to the central metal ion of a complex, the symmetry is disturbed much more markedly and a spectrum quite different from that expected for the free ion is observed. For example, even the strongest absorption band

at 1100 cm.$^{-1}$ characteristic of the free sulfate ion disappears completely in the complex $[Co(NH_3)_5\cdot SO_4]Cl$.

However, when the ligands are coördinated by an essentially electrostatic bond, the symmetry of the ion is practically maintained. Glycinometal complexes are good examples in which carboxylate frequencies (about 1600 cm.$^{-1}$) have been observed, indicating that the carboxylate resonance in the glycino ligand is maintained nearly as in the case of potassium glycinate. If the structure in these complexes were $-C\diagdown_{O-M}^{O}$, we should observe the characteristic C=O frequency (about 1700 cm.$^{-1}$), in place of the carboxylate frequency.

DEPARTMENT OF CHEMISTRY
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RECEIVED AUGUST 31, 1953

STRUCTURE OF RESERPIN
Sir:

Reserpin, the new sedative and hypotensive principle of *Rauwolfia serpentina* Benth. has been isolated for the first time by Mueller, Schlittler and Bein.[1] The m.p., optical rotation, characteristic bands in the ultraviolet and infrared spectra and elementary analysis were also reported; however, no empirical formula was assigned.

We have been able to establish the empirical formulas of Reserpin[2] and its degradation products. The alkaloid was isolated by chromatography of the Oleoresin fraction on acid-washed alumina. The repeatedly recrystallized material melted at 264–265° (dec.) and was dried for eight hours at 120° and 0.05 mm. for analysis. Calcd. for C_{33}-$H_{40}O_9N_2$: C, 65.11; H, 6.62; N, 4.60; OCH$_3$ (6), 30.59. Found: C, 65.31, 65.11, 65.00; H, 6.88, 6.83, 6.68; N, 4.79; OCH$_3$, 30.22. *Mol. wt.* Calcd. 608.67; found: 586 ± 20 (electrometric titration, pK'a 6.6 in 66% dimethylformamide); 610 (X-ray methods), no C-methyl. The ultraviolet spectrum of Reserpin showed the following bands: λ$_{max.}$ 216 mμ (log ε = 4.79), λ$_{max.}$ 267 mμ (log ε = 4.23), λ$_{max.}$ 295 mμ (log ε = 4.07), shoulder at 225 mμ, λ$_{min.}$ 246 mμ (log ε = 3.99), λ$_{min.}$ 286 mμ (log ε = 4.00). The maleate salt of Reserpin melted at 226–227° (dec.). Calcd. for $C_{33}H_{40}$-$O_9N_2\cdot C_4H_4O_4$: C, 61.31; H, 6.12; N, 3.86. Found: C, 61.5; H, 6.3; N, 3.83. Basic hydrolysis of Reserpin, using dilute sodium hydroxide in methanol yielded two fragments. One was readily identified as 3,4,5-trimethoxybenzoic acid, identical in each respect with an authentic sample (X-ray patterns, infrared ultraviolet, m.p. and mixed m.p.). The second fragment for which we propose the name Reserpic acid was isolated as the hydrochloride of an amino acid, m.p. 274–275° (dec.). It crystallized with one mole of methanol and was

(1) J. M. Mueller, E. Schlittler and H. J. Bein, *Experientia*, 8, 338 (1952).

(2) Our Reserpin was spectrally identical with a sample obtained through the courtesy of Dr. E. Schlittler of Ciba Pharmaceutical Products, Summit, N. J.