All samples of Tiberias water examined in this fashion yielded a large initial net alpha counting rate, which decayed within a few hours to levels close to background. The half-life obtained from a weighted mean of observations of 21 different samples was 36.8 ± 2 min.

It is well known that the "active deposit" of radon yields an alpha emission with a half-life of this order. The theoretical half-life of the "deposit" may be calculated from the known disintegration constants of Ra A, Ra B, and Ra C, but the result depends upon the relative initial quantities of these substances present in the sample pan at the moment it is introduced into the counter. Adopting reasonable values for these relative quantities, the theoretical half-life of the deposit may be shown to be 38.3 min, in good agreement with the experimental value quoted above. Hence, dissolved radon must account for the major portion of the radioactivity of Tiberias Hot Springs.

The concentration of radon in the water may be estimated on the basis of the initial net counting rates of the samples if corrections are made for: (*a*) the decay of the radon during the interval between bottling and preparation of the sample, (*b*) decay of the active deposit during the time (10 or 15 min) required to determine the "initial" count, and (*c*) self-absorption of the alpha particles in the sample. The modes of application of the first two corrections are obvious; the third correction is effected by means of the Bragg-Kleeman rule, as described by Evans and Goodman (*2*).

In the case of the strongest of the 21 samples examined, the initial alpha counting rate was 13.8/min, which, allowing for the background of 0.2/min, reduces to 13.6 net cpm. After application of the above three corrections this was found to correspond to a concentration of 2.18×10^{-11} curies/ml water from Tiberias Hot Springs. Most of the samples yielded an activity corresponding to at least one third this value; a few were as weak as one fifth.

A number of cold springs in the neighborhood of Tiberias were tested by the same method. The chief radioactive ingredient was again found to be radon, but the concentration was lower, ranging between 10^{-12} and 3×10^{-12} curies/ml water.

Tiberias Hot Springs were examined for traces of long half-life activity by means of aged samples of salts (obtained by evaporation of the water), which were introduced into the counter. In some cases no activity above background was detectable, even when very long counting times were resorted to; in others the net counting rate, corrected for absorption of the alpha particles within the sample, corresponded to as much as 10^{-12} curies of long half-life alpha activity/ml spring water. Apparently, some long half-life alpha emitter is present in Tiberias Hot Springs but, being a solid, it is naturally far less uniformly distributed than the radon. The average long half-life activity was found to correspond to as little as 10^{-13} curies/ml water. (This explains the results obtained by emulsions, referred to in the first paragraph.)

An attempt was made to identify the element responsible for the long half-life alpha emission by studying a few of the more active samples. Measurements of the range in aluminum of the alpha particles were carried out, but were necessarily inconclusive, owing to the very low activity and unavoidable thickness of the sources. They sufficed only to rule out elements such as uranium, with a very low energy spectrum.

Theoretically, the springs must contain some Ra D. The concentration of this long-lived isotope depends upon the length of time a specified concentration of radon is maintained, through an underground supply, in a given milliliter of water. This, in turn, depends upon unknown data, such as the manner in which the radon is picked up and circulated in the springs. Ra E and Ra F will, of course, be present in equilibrium with the quantity of Ra D. Thus, it is possible that all or part of the long half-life activity observed in aged Tiberias salts is due to the alpha emitter Ra F, maintained at a constant level by the decay of the 22-year half-life Ra D. Some experimental evidence supporting this theory was obtained, but was not entirely conclusive.

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An Action of ACTH on Adrenal Slices[1]

Robert Haynes, Kenneth Savard, and Ralph I. Dorfman

The Worcester Foundation for Experimental Biology, Shrewsbury, and The Department of Biological Chemistry, Harvard Medical School, Boston, Massachusetts

The studies presented in this communication demonstrate, by two separate methods, a stimulatory effect of added ACTH on corticosteroid hormone production by adrenal cortical slices under conventional incubation conditions. Since the completion of this work Saffran, Grad, and Bayliss (*1*) have reported that the addition of ACTH to rat adrenal halves *in vitro* results in an increased output of adrenal cortical hormones as measured by the ultraviolet absorption of extractable lipid material and by biological assay.

The procedure followed in this study was, with slight variations, as follows: Fresh beef or pork adrenals were sliced freehand or with a Stadie-Riggs microtome. The slices were divided into two groups and placed in citrated whole beef blood containing sodium acetate-1-C^{14}. To one group was added ACTH (approx 6 μg of an Astwood preparation per g tissue every 15 min); the other group served as a control. The incubations were for 2 hr at 37° C in an atmosphere of 95% O_2 and 5% CO_2. After incubation the slices and blood were extracted with 70% aqueous

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TABLE 1

INFLUENCE OF ACTH ON *in Vitro* INCORPORATION OF ACETATE-1-C^{14} INTO 17-HYDROXYCORTICOSTERONE

Acetate (μc/g gland tissue)	Compound F (17-hydroxycorticosterone)			
	Control		Added ACTH	
	Amount carrier F added (mg)	Specific radioactivity of isolated F (cpm/mg)	Amount carrier F added (mg)	Specific radioactivity of isolated F (cpm/mg)
13.5 (Hog)	0.5	46	0.5	92
83 (Cow)	2.0	102,000	2.0	200,000
94 (Bull)	2.15	3,370	2.15	7,350

ethanol, the extract was washed with ligroin, and the alcohol removed by distillation *in vacuo*. The remaining aqueous solution was extracted with methylene chloride, and the resulting extract was fractionated by chromatography on a column of silica gel. To each of the resulting corticosteroid fractions, a calculated weight of crystalline 17-hydroxycorticosterone (Table 1) was added as carrier. The diluted fractions were then chromatographed on filter paper strips in toluene-propylene glycol (*2*). The 17-hydroxycorticosterone zones were located, cut from the paper, eluted, and the specific radioactivity was determined (Table 1). Areas adjacent to the steroid zones were also eluted and were found to contain insignificant amounts of radioactivity. Characterization of the radioactive 17-hydroxycorticosterone eluted from the paper chromatograms was achieved by repeated chromatography on paper without decrease in specific radioactivity and by oxidation to adrenosterone. This oxidation of the 17-hydroxycorticosterone was carried out with chromic acid and gave rise to a ketosteroid which migrated on a paper chromatogram (ligroin-propylene-glycol system) as adrenosterone. The specific radioactivity of this steroid was of the same order as that of the 17-hydroxycorticosterone from which it was derived. The results of these experiments show that incorporation of C^{14} into 17-hydroxycorticosterone was increased twofold in the presence of ACTH.

Experiments were also done which demonstrated that added ACTH increased the output of formaldehydogenic steroids from adrenal slices. Corticoid extracts were chromatographed on silica gel, and the amounts of corticoids present were estimated by periodate oxidation of the appropriate column fractions, followed by determination of the liberated formaldehyde. Data from one such experiment given in Table 2 show that incubation with ACTH substantially increased the output of formaldehydogenic steroid by the slices. Liver slices, incubated and analyzed in the same fashion as the adrenal slices, showed no formaldehydogenic steroids before or after incubation.

TABLE 2

INFLUENCE OF ACTH ON *in Vitro* OUTPUT OF FORMALDEHYDOGENIC SUBSTANCES
(Values in μg formaldehydogenic steroid per g tissue)

Steroid initially present	Steroid at end of incubation						Output			
	Control			ACTH			Control		ACTH	
	No. vessels	Av	Range	No. vessels	Av	Range	Av	Range	Av	Range
44	4	71	62–78	4	126	107–148	27	18–34	82	63–104

In summary, the data indicated that the output of corticosteroids by adrenal cortex slices was substantially enhanced by the action of ACTH. The fact that ACTH stimulated the incorporation of C^{14} from acetate-1-C^{14} into 17-hydroxycorticosterone agrees with the finding of Hechter *et al.* (*3*) in perfused adrenals that ACTH accelerates the synthesis of corticosteroid hormones and not merely increases the rate of their release from the gland.

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Concerning the Presence of Citrate in Commercial Crystallized Bovine Serum Albumin[1]

Isaac Feldman and Jean R. Havill

Department of Radiation Biology,
School of Medicine and Dentistry,
University of Rochester, Rochester, New York

It has become a widespread custom to use Armour's crystallized bovine serum albumin, without pretreatment, as a representative protein in the study of the theory of ion-protein binding. This preparation is obtained from citrated blood by the procedure of Cohn *et al.* (*1*). It appears to be generally assumed that all but an insignificant amount of the citrate is removed by the crystallization process. However, during the course of an investigation of the binding of beryllium by albumin, it has been discovered that the commercial albumin preparation contains a significantly large amount of a strong complexing agent. Considering the relative binding strengths of the various possible contaminants, it is believed that this anion is citrate and is present to the extent of at least 0.5 moles/mole protein.

Beryllium begins to hydrolyze and polymerize near pH 4.6, even in concentrations as low as 10^{-9} *M* (*2*).

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