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Manuscript received November 15, 1951.

A Saliva Test for Prenatal Sex Determination

Gustav Wm. Rapp and Garwood C. Richardson

Department of Biochemistry, Loyola University School of Dentistry, and 30 N. Michigan Ave., Chicago, Illinois

During the course of investigating some of the many ramifications of the Richardson Pregnancy Test (*1*) two rather interesting observations were made. The Richardson test depends upon the presence of free estrone, in contrast to bound or modified estrone or similar 17-ketosteroids in the female urine. This level of free estrone substances rises sharply soon after conception, the test being positive in as little as 2 weeks after conception, sometimes even before the first missed menstrual period. We were naturally interested in determining whether the free estrone level rose in other body fluids. Blood and urine have already been mentioned in Richardson's original article. We investigated such fluids as saliva, tears, and perspiration. The studies upon saliva yielded the results that are the subject of this report.

It was noted early in the study that in only some of the women who were in their sixth or seventh month of pregnancy did the Richardson test prove positive when the saliva was tested. In each of these cases, however, the test was positive on the urine. The apparent answer to the problem was forthcoming only after the delivery of the child. In nearly every case, the positive tests were associated with a male child, and most of the negative tests were associated with a female child. A detailed study followed. The results are presented in Table 1.

TABLE 1

RELATION BETWEEN SEX OF CHILD AND REACTION OF THE MOTHER'S SALIVA TO THE RICHARDSON TEST

	Positive	Negative
Males	218	3
Females	7	148

The precise nature of the substance responsible for the positive test is not known. It is believed that some androgenic substance is being identified, since, whereas a nongravid female normally yields a negative test, after the injection of testosterone or androsterone a strongly positive reaction results. The male saliva, spermatic fluid, and blood serum are all strong positive reactors.

The selective excretion of certain blood constituents through the salivary gland is well known. These findings illustrate a rather delicate selectivity of female salivary glands in their capacity to screen out certain female-associated hormones, but to allow certain male-associated ones to pass into the salivary fluid.

A detailed report of this project will be published in suitable medical journals. Because of their obvious practical nature, these findings are preliminarily reported here to allow early verification by others.

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Manuscript received October 15, 1951.

Influence of Cobalt on Reproduction of Mice and Rats

Werner G. Jaffé

Instituto Nacional de Nutrición, Caracas, Venezuela

Reproduction studies with mice and rats kept on whole plant rations have been in progress in this laboratory for several years. In the first experiments, poor reproduction performance was observed (*1*), but later much more satisfactory results were obtained with a slightly modified diet (*2*). The only difference in the composition of the diets was that the salt mixture supplement used originally did not contain cobalt, whereas in the later experiments 0.2% $CoCl_2$ was added to the salt complement. Because of the apparent influence of this small amount of dietary Co on reproduction, the present study was undertaken.

The rats used were of the Sprague-Dawley strain, raised in our laboratory; the mice were of the same albino strain used in the previous experiments. The rats had been kept on a whole plant ration for at least 3 generations prior to the start of the present experiments, and the mice for at least 5 generations. The animals were kept on the basal ration in common cages until females became pregnant. These were put in single screen-bottomed cages and given the respective experimental diets and water *ad lib.* Litters were reduced by random selection to 6 and weaned at the age of 28 days.

The composition of the basal diet was: solvent-extracted soybean meal, 46; cornmeal, 46; sesame oil containing 0.2% percomorphum oil and 0.2% wheat germ oil, 5; salt mixture II USP, 2; thiamine, 0.3 mg; riboflavin, 0.3 mg; calcium pantothenate, 2 mg; pyridoxin, 0.2 mg; choline hydrocloride, 100 mg; nicotinic acid, 2 mg; folic acid, 0.025 mg; biotin, 0.01 mg; inositol, 10 mg; PABA, 25 mg. The diet has been analyzed by microbiological methods and found to contain about 0.5 μg/100 g of vitamin B_{12} activity (*2*). Cobalt was supplied in the diet by adding 0.2% $CoCl_2$ to the salt mixture, or in the drinking water in a concentration of 0.2 mg% of $CoCl_2$; when cobalt was injected, a 0.1% solution of $CoCl_2$ in physiological saline solution was used, of which 0.2 ml was injected in each female 2–6 days prior to giving birth. A total of 130 litters of mice and 64 litters of rats was used in the present study.