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## THE EXCRETION OF PHENOL RED BY THE FROG, *RANA CATESBIANA*

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In an earlier paper it was demonstrated that inulin and creatinine are excreted by the frog kidney solely by the process of ultrafiltration at the glomerulus, neither substance being secreted nor reabsorbed by the renal tubule cells (Forster, 1938). In this study of 92 clearance periods in 14 animals, simultaneous inulin and phenol red determinations are made in order to analyze the processes involved in the renal elimination of the latter. In all marine and terrestrial animals investigated, in which quantitative examination has been possible, it has been shown that tubular excretion plays a part in the elimination of phenol red, and that this process is limited by a maximal rate (see Shannon, 1939). However, on the basis of direct analysis of the glomerular filtrate in the frog, Richards and Walker (1935) concluded that all the phenol red excreted might have been contained in the glomerular filtrate. In this clearance study of phenol red elimination the difference between the total amount of phenol red excreted and the calculated amount filtered at the glomerulus indicates that a large fraction of the phenol red is excreted by the renal tubule cells, and that this tubular fraction has a maximal value such as has been demonstrated in the marine and terrestrial animals.

The methods used in this study for inulin determination and the collection of blood and urine were the same as those employed in an earlier investigation (Forster, 1938). Usually 2 grams of inulin and varying amounts of phenol red, depending upon the desired plasma concentration, were injected into the dorsal lymph sac 3 to 4 hours before the first urine collection. Hourly clearances of inulin and phenol red were then determined on a falling plasma curve obtained by analyses of blood collections taken at the beginning, middle and end of a 5 to 12 hour experiment. A photoelectric colorimeter was used for many of the low phenol red plasma and urine concentrations, and a Duboscq-type microcolorimeter with a 74 Wratten filter, for the others. An average plasma blank determination was obtained (0.167 mg. percent) and this value was subtracted from all plasma phenol red readings.

The free phenol red fractions at various total concentrations in the plasma were determined from pooled samples of plasma taken from 3 animals after the method of Shannon (1935). These values vary from 45 percent free at very low plasma total phenol red concentrations, to 74 percent free at high levels (112 mg. percent). The filtration rate of phenol red elimination was determined as the product of the inulin clearance and the percent free of plasma phenol red,

without reference to the protein or water content of the plasma. The rate of tubular excretion was determined by subtracting the filtration rate from the total rate of excretion (UV).

As has been shown with all substances excreted by tubular activity, the phenol red/inulin clearance ratio is depressed as the phenol red plasma concentration increases in the frog. These values range from 5 to 10 with a phenol red plasma concentration of about 1 percent, and fall below 1.0 when the concentration is higher than 40 mg. percent, thereby emphasizing the unavailability of the plasma protein-bound fraction for filtration. No attempt was made to obtain a maximal phenol red clearance because of the difficulties involved in obtaining accurate phenol red plasma determinations of less than 1.0 mg. percent. The maximal phenol red clearance probably lies above 185 ml. per kg. per hr. (the average inulin clearance in this experiment was 27 ml. per kg. per hr.). At phenol red plasma concentrations of 2.0 mg. percent the average phenol red clearance is 80 ml. per kg. per hr. and the average phenol red/inulin clearance ratio 4.2.

Calculations of the rate of tubular excretion show that a tubular secretory maximum is reached with a total plasma phenol red concentration of about 7.0 mg. percent. The maximum amount of phenol red which the frog renal tubules can transport from peritubular fluid to lumen is roughly 0.50 mg. per kg. body weight per hr. or 13 mg. per kg. per day.

#### Summary:

1. The process of tubular excretion is a large factor in the renal elimination of phenol red in the frog.
2. The rate of tubular excretion reaches an apparent maximum at a total phenol red plasma concentration of 7.0 mg. percent.
3. The maximum amount of phenol red which the frog renal tubules can excrete is approximately 0.50 mg. per kg. body weight per hr.

#### REFERENCES

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## THE MORPHOLOGY OF THE DOGFISH RENAL TUBULE

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In connection with a study of the process of acidification of the urine, and to lay a groundwork for other studies of a similar nature, an examination of the structure and configuration of the kidney tubule of *Squalus acanthias* has been undertaken. While this is still incom-