

# Experiment HK-1: Human Kidney

## Background

The mammalian kidney plays a major role in waste excretion and the balance of water and electrolytes. This role in osmoregulation will be examined in this laboratory session.

Changes in the osmotic state of bodily fluids occur daily as we work, play, eat, drink and sleep. These alterations are minimized through feedback control mechanisms which allow osmoregulatory organs to adjust their activity and maintain the stability of the internal environment. A stable internal environment buffers the body's tissues against the variations and extremes of the external environment.

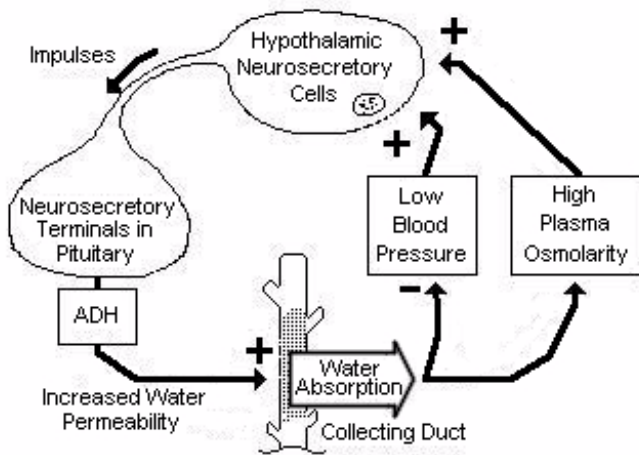


Figure HC-4-1: Feedback regulation of blood osmolarity by action of ADH.

The control of water retention by the human kidney is a well known example of one of these feedback regulations (Figure 1 on page 1). This particular system involves both neural and endocrine mechanisms. Trace the process with the following example. The body perspires to release heat and cool off, but it also releases water as evaporation occurs on the surface of the skin. The lost water is replenished from a concentration gradient through adjacent tissues and ultimately from the circulatory system. Unless the water lost from the circulatory system is replaced quickly enough by water that is ingested, the plasma osmolarity will increase. Osmotically sensitive neurons with cell bodies located in the hypothalamus respond to the increased plasma osmolarity with an increased rate of firing. The impulses are carried down the axons of these cells to terminals located in the posterior lobe of the pituitary gland, the neurohypophysis. The hormone, ADH (Antidiuretic Hormone), is released from the terminals of these neurosecretory cells to the surrounding circulatory

system. The increased neural activity of these cells causes an increased release of ADH. ADH travels through the circulatory system to its target tissue, the epithelial walls of the collecting ducts in the kidney.

The walls of the collecting ducts become more permeable to water as the blood level of ADH increases. With the increased permeability of the collecting duct walls, more water is osmotically drawn out of the collecting duct into the surrounding interstitial fluid. The water flows in this direction because the fluids and tissues of the renal medulla that surround the collecting ducts are made hyperosmotic by the countercurrent mechanism of the Loop of Henle. So, as ADH increases the permeability of the collecting ducts and more water is drawn out of the urine as it passes down the duct toward the renal pelvis, more water is conserved by the body and a urine more concentrated in solutes is created.

The water conserved by the collecting ducts reenters the circulatory system through the peritubular capillary network that surrounds the entire tubular system of the kidney. The plasma osmolarity decreases as the solutes in the blood are diluted by the reabsorbed water. The lower plasma osmolarity is sensed by the osmotically sensitive cells in the hypothalamus and the firing rate of these cells is reduced. Thus, the secretion of ADH is reduced and the permeability of the collecting duct walls to water is reduced until another increase in the plasma solute concentration is detected.

## Preparation

- 1 Do not drink anything but water during the 3 hour period before lab. You may drink as much water as you wish.
- 2 Urinate one hour before coming to lab. Do not urinate again until the first collection at the beginning of laboratory.
- 3 If you have circulatory problems, poor kidney function or have any medical problems, do not volunteer as a subject for this experiment.

## Equipment Required

- Drinking cups
- Urine collecting cups
- Urine Hydrometer

## Exercise 1: Measuring Urinary Output

Aim: To drink a particular solution and measure the amount of urine produced over time.

### Procedure

- 1 At the start of laboratory, obtain a couple of graduated urine specimen cups and collect the entire contents of your bladder.

- Note the time when this collection was made, the time of your last urination before lab, and the volume of urine collected.
- Calculate the control rate of urine production (ml/min) by dividing the urine volume (mls) collected by the number of minutes since your last urination.
- You will be assigned to drink one of the following solutions (Table HC-4-1 on page 2). These assignments will be made at random. If you have any medical condition which prohibits you from consuming one of these solutions, inform the lab staff. You can pick another solution or be excused from being a subject.

**Table HC-4-1: Solutions used in Kidney Experiment**

Solution	Dosage
Drinking water	When thirsty (Record time between drinks)
Drinking water	16 ml per kg of body weight
Drinking water	7.5 ml per kg of body weight
Cola	7.5 ml per kg of body weight
Gatorade	7.5 ml per kg of body weight
Caffeine-free Cola	7.5 ml per kg of body weight
Diet Caffeine-free Cola	7.5 ml per kg of body weight

- If you are drinking any solution at the rates of 7.5ml/kg or 16ml/kg body weight, consume the total volume of the solution as quickly as possible.
- Collect urine samples at 30, 60, 90, and 120 minutes after drinking the solution. If you need to urinate more frequently, keep a record of the total volume of urine collected during each 60 minute interval.

### Measurements

#### URINE PRODUCTION RATE (ML/MIN)

- Use a graduated specimen cup to measure the volume of urine collected at the 0 (control), 30, 60, 90, and 120 minute sampling times.
- Calculate the urine production rate in milliliters per minute for each of the four sampling times.

#### URINARY SPECIFIC GRAVITY

- For each time point (0, 30, 60, 90, 120 minutes) retain enough of your urine sample to fill 80% of the hydrometer cylinder.
- Place the urinometer in the chamber and gently spin the device so that it rotates freely for at least two turns.
- Read the specific gravity of the urine sample by matching the meniscus of the sample with scale on the stem of the urinometer. The scale ranges from 1.000 at the top to 1.060 at the bottom.

- If the specific gravity of a urine sample is too high (>1.060) or there is an insufficient volume of urine collected, measure a given volume of urine and dilute that volume by adding one or two times more distilled water.

- Account for the dilution when the specific gravity of the urine is recorded. For example, if one volume of water was added to one volume of urine and the urinometer read 1.02, specific gravity would be calculated by:

- Subtracting 1.000 (specific gravity of water) from 1.020 = 0.02;
- Multiplying the difference (0.02) x 2 (for 2 volumes) = 0.04; and
- Adding 0.04 to 1.000 = 1.04, the specific gravity of sample.

#### OPTIONAL MEASUREMENTS: PH, URINARY GLUCOSE

- pH:** Use assorted pH papers to determine urine pH of the samples taken at the 0, 30, 60, 90, and 120 minute sampling times.
- Urinary Glucose:** Use Diastix (tm) to check for the presence or absence of urinary glucose. Follow the directions for the use of Diastix indicator strips as listed on the container.

#### Data Analysis

- Pool the calculated data from all the subjects in the class. Add your urine production rate and specific gravity data for each time point to a large class table. The data from subjects who consumed the same solution should be grouped in the same area of the table.
- Calculate the average urine production rate and average specific gravity for each time point for all subjects in the group who drank the same experimental solution. Discard any obviously erroneous results.
- Make line graphs of your urine production rate and specific gravities as a function of time.
- Examine the results and graphs from other subjects who drank different solutions.

#### Questions

- Compare the different groups who drank 7.5ml of fluid for each kg of body weight. Which group had the highest average urine production rate? Why?
- Compare the different groups who drank 7.5ml of fluid for each kg of body weight. Which group had the highest average specific gravity? Why?
- Compare the groups who drank 7.5ml and 16 ml of water for each kg of body weight, and water at will. Which group had the highest average specific gravity? Which group had the highest average urine production rate? Why?
- During the experiment, which group probably had the highest concentration of ADH in their bodies?