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Examination of Urine

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Urine is the end waste product of the body obtained through filtration of blood. Renal diseases, diabetes mellitus, urinary calculi in kidneys or at other places in tract, jaundice, ascites, edematous conditions of body, hematuria and hemoglobinuria. The examination of urine is not only helpful in concluding the diagnosis of disease but also useful in assessing the prognosis of condition.

Collection

The urine samples can be collected by following methods.

1. In large male animals, the polythene bags can be applied over the urethra and tie them with the help of rope over the back. When animal urinates the urine is collected in the polythene bags which should be removed from animals at different intervals. It is the method of choice for quantitative examination of the urine sample.
2. In cows, buffaloes, sheep and goat, one can stimulate the urethra through vulva, which allows the animal to urinate.
3. In large animals, urine can also be collected by applying pressure on urinary bladder through rectum.
4. In small animals, urine can be collected directly from the bladder through syringe and needle using aseptic conditions.
5. In some animals, urine is collected by catheterization.

Preservation

Normally the urine sample is examined just after its collection. However, when delay is expected, it should be kept in refrigerator at 4°C without adding any preservative. For a longer storage, the toluene can be added in urine to form a layer over urine; it suits for chemical examination. One drop of 40% formalin can also be used as preservative in urine but it may give false reaction for sugar examination. The addition of thymol 0.1 gm/ 100 ml of urine is also recommended but it may give false reaction for albumin contents. One drop of phenol per 10 ml of urine can be used as preservative without much deterioration. The best way is to keep the urine sample at 4°C in refrigerator without addition of any preservative. All attempts should be made to examine the urine sample as soon as after its collection for proper interpretation.

Examination

Physical Examination

Colour

- The colour should be noted at the time of urine collection, by keeping it in a glass tube. Normally the colour of urine is water colour to amber colour. However, in certain disease conditions the colour of urine alters:
- Hematuria: Red colour urine, cloudy.
- Hemoglobinuria: Brown to red colour.
- Icterus: Yellow brown colour to greenish yellow.

- Pyogenic infections of kidneys: Yellowish green colour.
- High fever/dehydration due to diarrhea/ vomiting: Yellow orange to reddish colour.
- Azoturia in horses: Brown to black colour of urine.
- Phenothiazine, phenolphthalein and/or azosulfamide treatments: Red colour urine.

Odour

The normal odour of urine is uremic. Ketosis, the odour of urine is sweetish or fruity while it becomes fetid in pyogenic infections of kidneys. The abnormal sample may give the odour of ammonia, acetone or fetid odour.

Turbidity

Normally the urine of equines is turbid and cloudy due to calcium carbonate and mucous while the urine of other animals is clear and transparent. Yellowish turbidity can be observed in pyogenic infections of kidneys. Kidney damage may be responsible for hema- turia, albuminuria and proteinuria, which gives a turbid urine. Cloudiness in urine is due to presence of leukocytes, erythrocytes, epithelial cells, bacteria, mucous, fat and/or crystals. Of these, leukocytes give a white cloud while the red turbidity is seen due to erythrocytes and epithelial cells. Bacteria forms a uniform cloudiness in urine and fat forms clouds, on addition of chloroform or ether in the urine sample and urine becomes transparent. On centrifugation, the, fat comes at the top of urine while other turbid material settles down.

Foaming

On shaking the normal urine produces white foams but in case of proteinuria, the amount of foam is in excess which remains for a longer duration.

Specific Gravity

The specific gravity of urine is measured by urinometer. The urine is filled in cylinder and urinometer is left in the urine which should not touch the walls of cylinder. The specific gravity increases in acute interstitial nephritis, cystitis, diabetes mellitus and dehydration and it decreases in chronic interstitial nephritis, pyometra and diabetes insipidus.

Chemical Examination

pH

Requirements pH strips or pH meter

Animals Normal pH

Ruminants 7.4-8.4

Canine 6-7

Horses 8.0

Interpretation

Urine becomes acidic during starvation, fever, treatment of ammonium chloride, sodium chloride, calcium chloride and sodium acid phosphate. The alkaline urine occurs in cystitis and due to treatment of acetate, bicarbonate, citrate and nitrates of sodium or potassium.

Glucose

Requirements Benedict's reagent, test tube

Procedure

1. Take 0.5 ml of urine in a test tube.
2. Add 5.0 ml of Benedict's reagent and mix them thoroughly.
3. Place this tube on a boiling water bath or flame for 5 minutes.
4. Remove the tube from heating and keep on test tube stand for few minutes and note the change of colour.

Blue (–) No sugar

Blue to green sediment + mild sugar

Yellow with heavy sediment ++ moderate

Orange with heavy sediment ++++ highly positive for sugar.

Interpretation

Glycosuria occurs due to hyperglycemia and in diabetes mellitus, acute or chronic pancreatitis, hyperthyroidism, hyperadrenalism, hyperpituitarism, increased intracranial pressure,

enterotoxemia, and certain drugs like streptomycin, chlortetracyclin, penicillin, tetracyclin and chloramphenicol may also lead to glycosuria.

Protein

Requirements Robert's reagent

Procedure

- Take 2 ml urine in a test tube and place 2 ml Robert's reagent over the urine.
- If the albumin is present in the urine, the white ring will appear at the interjunction of two fluids.

Interpretation

• Very heavy ring	++++	Highly positive
• Heavy ring	+++	Positive
• A wide ring	++	Moderately positive
• A narrow ring	+	Mild positive
• No ring	(–)	Negative

Proteinuria

Excessive muscular exertion, stress, more protein in diet and convulsions which may disappear after removal of the factor. In disease conditions, proteinuria may occur in nephritis, acute interstitial nephritis, pyelonephritis and nephrosis. Phenol, arsenic, lead, mercury, sulfonamides, phosphorus, turpentine and ether may cause proteinuria. Proteinuria may also occur in cystitis, prostatitis, pyelitis, urethritis, ureteritis, urolithiasis and due to mixing of vaginal and prepucial discharges in the urine.

Ketone Bodies

Requirements

sodiumnitropruside powder, ammonium sulfate, 20% ammonium hydroxide solution.

Procedure

1. Mix 1.0 gm of sodiumnitropruside powder to 100 gm ammonium sulfate powder thoroughly and place 1 gm mixed powder in a test tube.
2. Add 5 ml urine and mix properly.
3. To this overlay slowly 20% ammonium hydroxide solution.
4. In positive case, the colour becomes red to purple at the interjunction of two fluids.

Interpretation- Bovine ketosis, pregnancy toxaemia of ewes, pancreatitis, hepatitis

Bile Salts

Hey's test

Requirements: Sulfur granules, test tube

Procedure

1. Take about 4-5 ml urine in a test tube and shake it.
2. Sprinkle sulfur granules over the surface of urine in test tube.
3. If urine contains bile salts, the sulfur granules sinks through urine.

Interpretation- jaundice, hepatitis, pancreatitis

Indican

Requirements-0.2% ferric chloride solution, chloroform,

Procedure-

1. Take 5 ml urine in a test tube
2. Add 5 ml 0.2% ferric chloride solution and 2 ml chloroform.
3. Mix well by inverting the tube and allow it to stand for some time.
4. The chloroform settles, examine the colour of chloroform.
5. The presence of blue to violet colour is indication of presence of indican.

Interpretation

The indican level is increased in intestinal obstruction, constipation, rapid breakdown of protein, indigestion, gastritis, enteritis, intussusception and volvulus.

Blood

Requirements- saturated benzidine solution, 3% hydrogen peroxide

Procedure

- Take 2 ml urine in a test tube and in another tube 1 ml of saturated benzidine solution and 1 ml 3% hydrogen peroxide and mix well.
- 2. The contents of the both tubes are mixed thoroughly.
- 3. If the blood is present in urine, it is indicated by the development of a green to deep blue colour.

Interpretation

The presence of blood in urine is known as hematuria which occurs in pyelonephritis, ureteritis, cystitis, urolithiasis, pyelitis, trauma in urethra, prostatitis, kidney neoplasms, acute nephritis, parasitic infection**.** If whole urine is mixed with blood the possible source may be kidneys but if only last portion of urine is red and containing blood, it is from bladder.

Hemoglobin/Myoglobin

Requirements- ammonium sulfate, test tube

Procedure

1. Take 5 ml urine in a test tube and add 5gm ammonium sulfate, shake well and allow to stand for few minute.
2. If the colour of urine becomes clear, it indicates the presence of hemoglobin in urine.
3. If the colour remains the same, as before the test, it is myoglobin.

Interpretation

Hemoglobinuria occurs in leptospirosis, piroplasmosis photosensitization, post parturient hemoglobinuria and bacillary hemoglobinuria.

The myoglobinuria occurs in azoturia.

Microscopic Examination

1. For microscopic examination, take 5-10 ml urine in a centrifuge tube and centrifuge it at 1000 rpm for 10 minute.
2. Discard the supernatent and place a drop of sediment on middle of the clean and dry glass slide.
3. Cover it with a coverslip and examine under microscope for epithelial cells, leukocytes, erythrocytes, casts, bacteria and crystals.
4. If the cells are not clear, the smear can be made from sediment and stained by Giemsa.
5. The colour of sediment becomes red if red blood cells are present in urine while it becomes yellow if bile pigments are present in urine. The white colour sediment is seen in the presence of crystals in urine.

The urine sediment is of two types,

Organized sediment: It contains epithelial cells, leukocytes, erythrocytes, microorganisms, casts, parasites.

Unorganized sediment: It includes fat, crystals, pigments, etc.