

Pregnancy Test

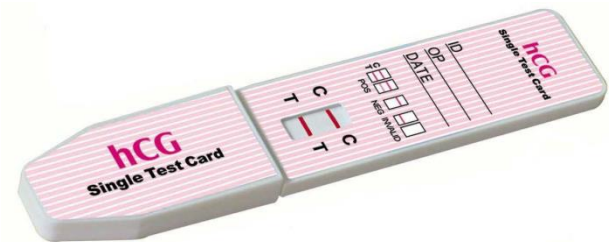
Detecting Human chorionic gonadotropin in urine

BCH 472



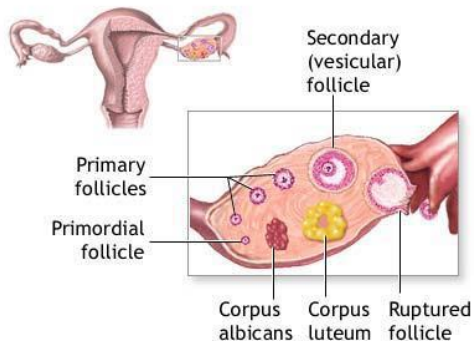
Human chorionic gonadotropin

- Human chorionic gonadotropin (hCG) is a glycoprotein hormone produced by a portion of the placenta following implantation.
- The **qualitative hCG test** can be used to see if a woman is pregnant or not.



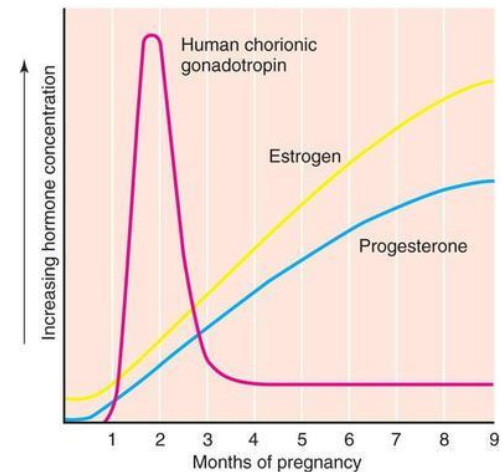
hCG Role in pregnancy:

- Promotes the maintenance of the corpus luteum during the beginning of pregnancy in the ovary . This allows the corpus luteum to secrete the **progesterone** during the first trimester. Progesterone enriches the uterus with a thick lining of blood vessels and capillaries so that it can sustain the growing fetus.
- Human chorionic gonadotropin also plays a role in cellular differentiation/proliferation.

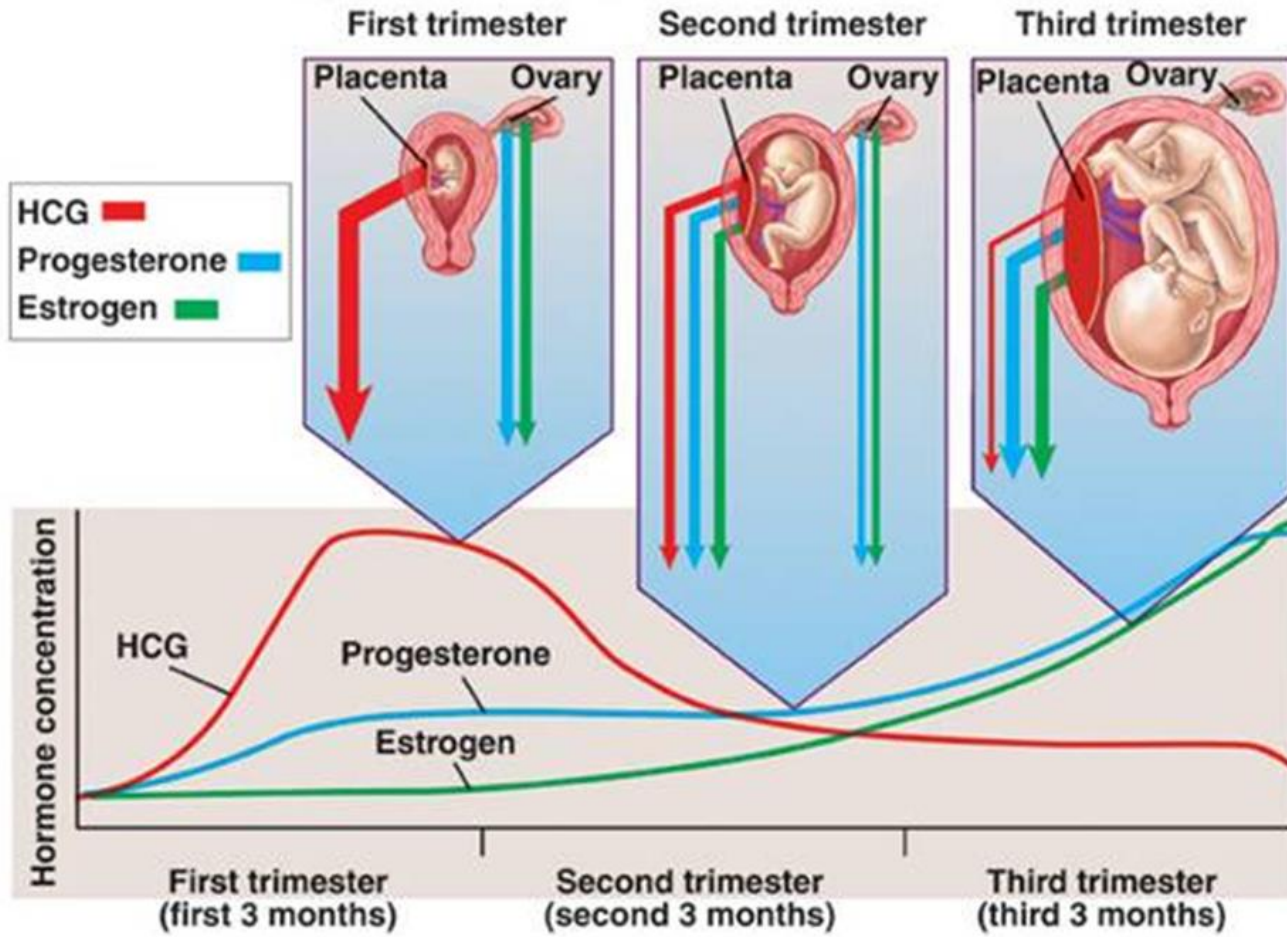


hCG level

- **During the first trimester**, hCG levels rise steadily and rapidly, peaking around 10 weeks' gestation, and subsequently taper off to less than 10% of peak levels.
- By the end of the third month it has reached a low level which will remain constant for the duration of the pregnancy.



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Notes:

- In pregnant women, depressed urine hCG levels may indicate threatened **abortion** .
 - Some cancerous tumors produce this hormone; therefore, **elevated** levels measured when the patient is **not pregnant** can lead to a **cancer diagnosis**.
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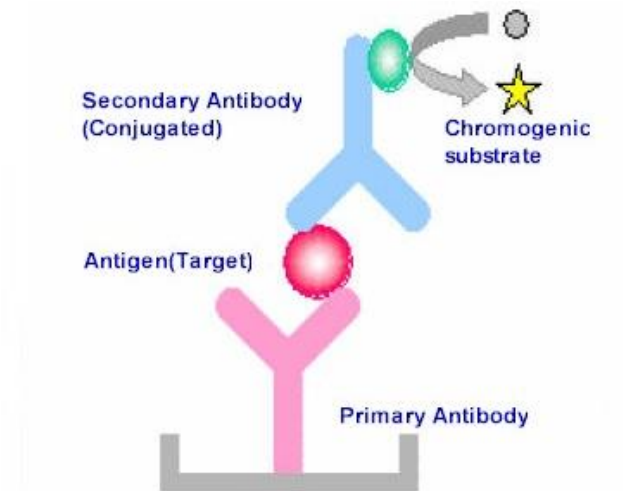
Practical Part

Objective:

To detect and confirm pregnancy.

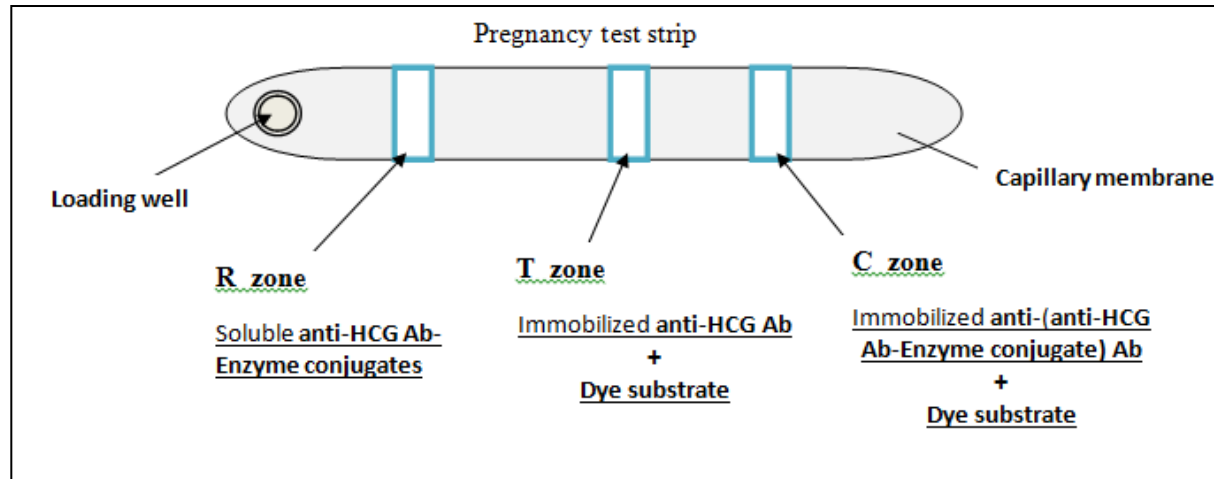
Principle:

- Urine pregnancy tests use the enzyme-linked immunosorbent assay (**ELISA**) technique, using a highly specific monoclonal antibody directed against the -subunit of human chorionic gonadotropin (-hCG).

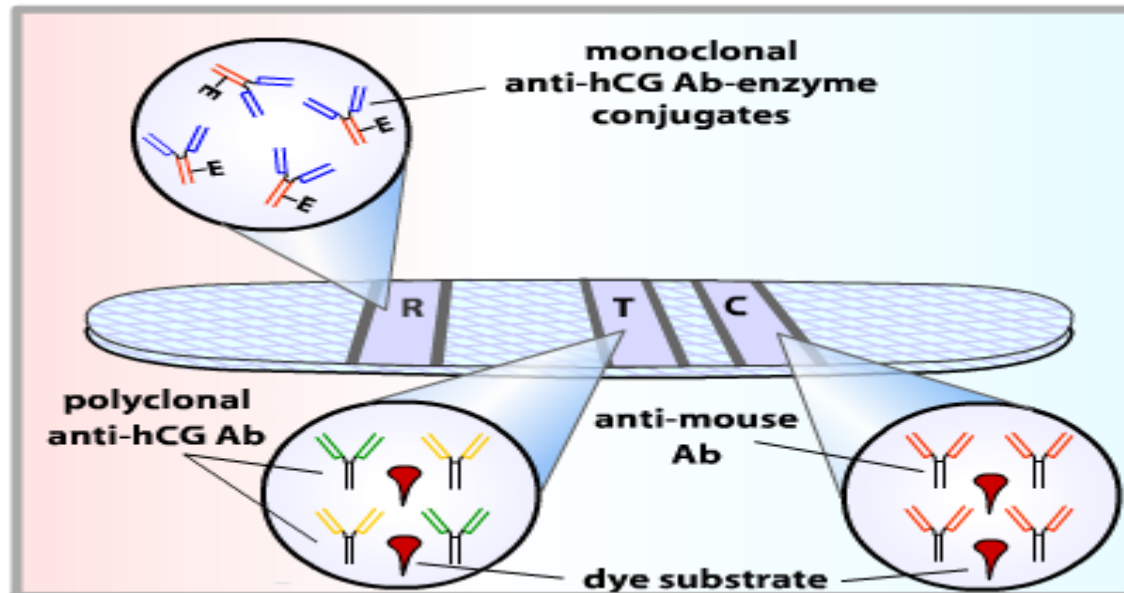


Principle:

Pregnancy test strip consist of :



- **The reaction zone:** contains soluble **anti-hCG antibody-Enzyme**. These are mouse monoclonal antibodies linked to an enzyme.
 - **The test zone:** contains immobilized polyclonal mixture of hCG- antibody + dye substrate.
 - **The control zone:** the dye substrates + anti-(anti-HCG Ab-Enzyme conjugates) Ab can recognize epitopes on the mouse monoclonal .
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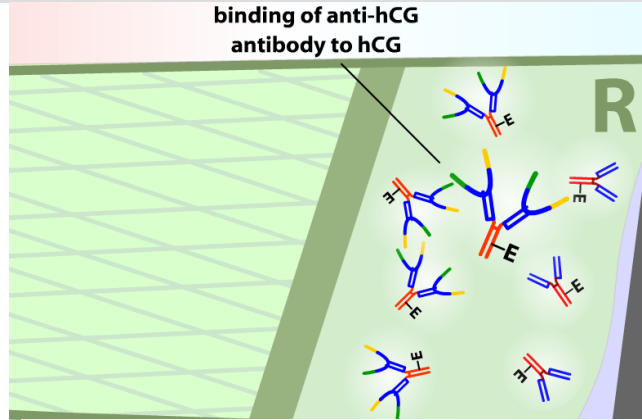
- <http://www.sumanasinc.com/webcontent/animations/content/pregtest.html>

Sequence of events:

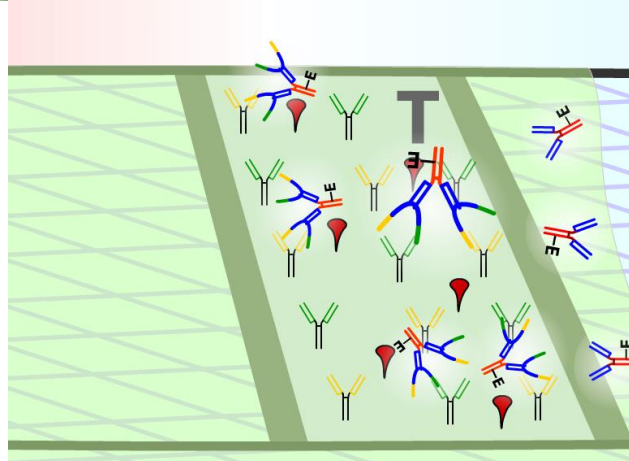
- A few drops of urine is transferred to the specimen well (loading well).
 - Urine will flow by capillary action from loading well towards zone R carrying along with it the HCG hormone.
 - **At R zone**, the HCG hormone will react and bind with the soluble anti-HCG Ab-enzyme conjugates forming a complex of **HCG hormone - HCG Ab - enzyme conjugate**.
 - This complex will migrate towards zone T.
 - **At zone T**, this complex will react and bind with the immobilized anti-HCG Ab, once it binds with the immobilized Ab, this will activate the enzyme thus allowing to act on the dye substrate and produce a **color that indicates a positive pregnancy result**.
 - The **excess** free HCG Ab - enzyme conjugates will pass from zone T to zone C.
 - **At zone C**, this free HCG Ab - enzyme conjugates, will react and bind with the immobilized anti-(anti-HCG Ab-Enzyme conjugates) Ab there, once bound it will activate the enzyme, thus allowing to act on the dye substrate and produce the **color** detecting at zone C which is an **indicator of the activity or reliability of the test**.
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binding of anti-hCG
antibody to hCG

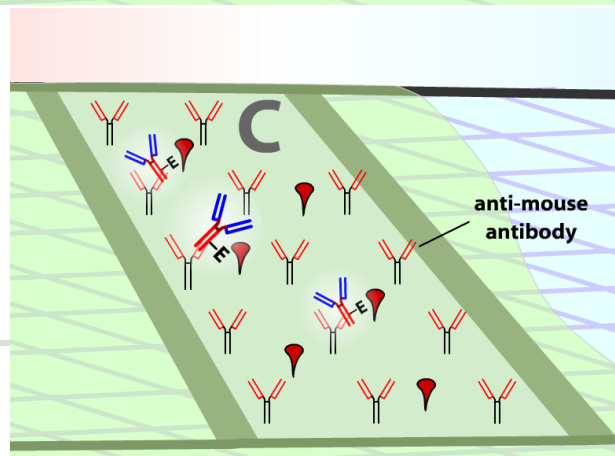
At R zone

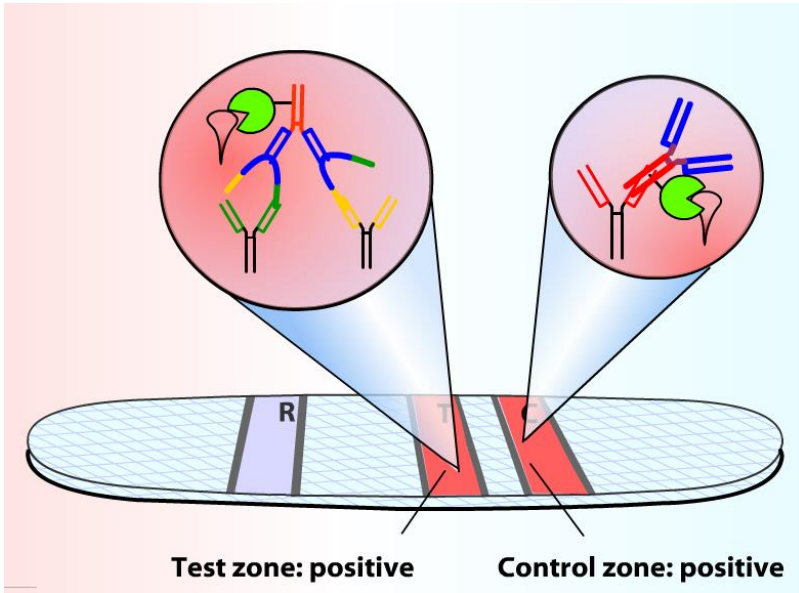


At T zone

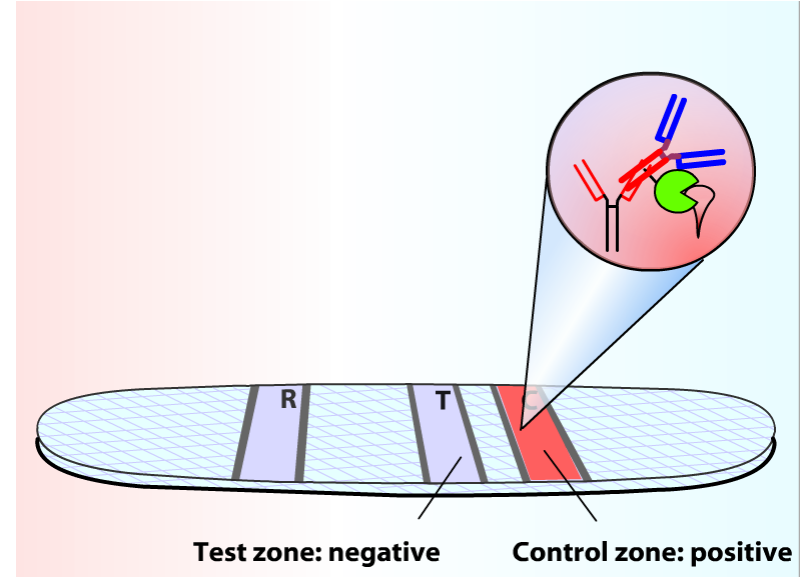


At C zone





Pregnant (+)



Non pregnant (-)

Specimen Collection and Preparation

- Collect at least 1 mL of urine in a clean, dry, plastic or glass container with no preservatives.
- Specimens may be collected at any time of the day, however the **first morning** sample generally has the **highest concentration** of HCG and is the specimen of choice.

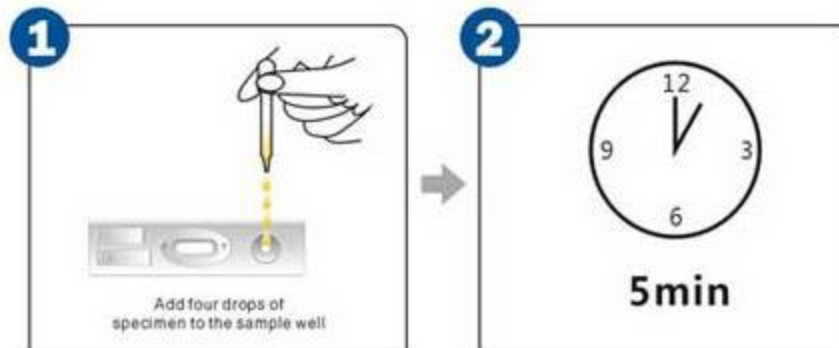


Procedure:

- Holding a Sample Dropper vertically, add exactly four drops of the urine specimen to the sample well.
 - Read results at time indicated in procedure.
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Test Procedure

Urine LH Cassette



Result Reading



Positive



Negative



Invalid

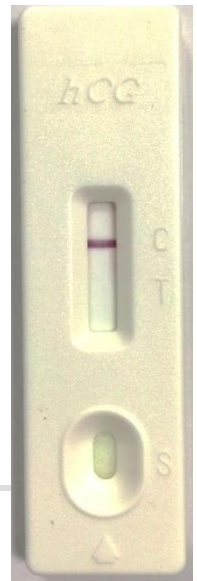
Results

- Follow the instructions on the reagent package insert provided by the instructor to properly perform the test.

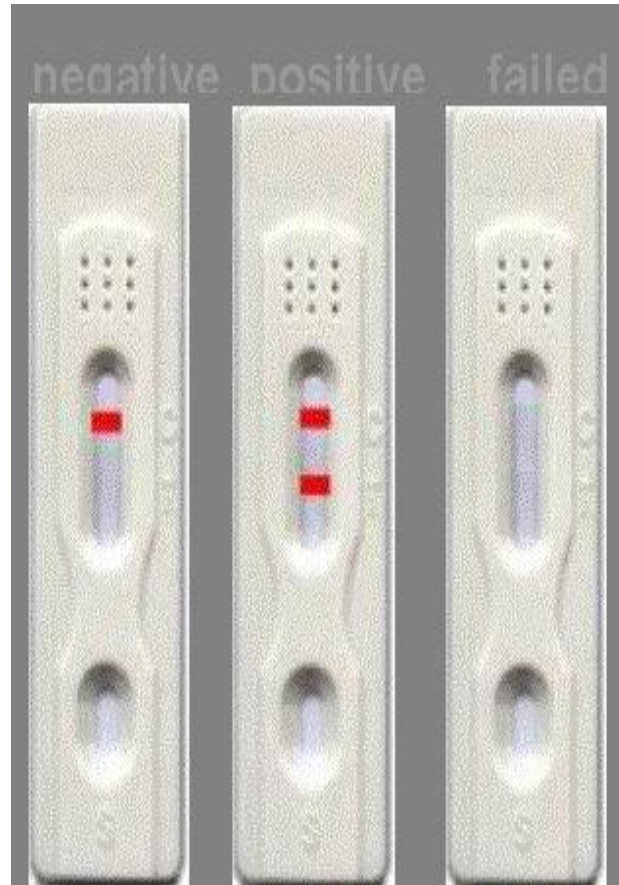
Interpretation of Results:

- Based on the package insert correctly interpret the results of the pregnancy test on the patient samples
- Record results as “**Positive**” or “**Negative**”

SAMPLE TESTED	RESULT
Urine sample	



Pregnancy test strip



Discussion

Comment on the results and state if sample is pregnant .
