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Fructose (Quantitative)

bio-marker of seminal vesicle activity

REF SP/SFT/F-012-B



IVD

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product



Turnaround time for test: 75-80 min



Store at: 2°C - 8°C after receiving

Fructose

biomarker of vesicle activity

CONCEPT

Poor quality semen may result from abnormal sperm production in testis, post testicular sperm damage in epididymis or abnormal ejaculate from accessory glands.

Seminal plasma receives contribution from seminal vesical, prostate & accessory glands. Seminal vesical secretions contribute 2/3 volume. Its main ingredients are fructose, prostaglandin & coagulating proteins. The normal function of the seminal vesicle is important for fertility. parameters as motility, sperm chromatin stability & immune protection are affected in case of hypofunction of seminal vesicles. Seminal vesicle secretions also protect sperm from ROS present in semen produced by leukocytes or sperm itself.

Fructose is the major energy substrate of sperm metabolism (Glycolytic, anaerobic) and is important for the maintenance of sperm motility.

Fructose serves as a bio-marker for seminal vesicle function.

1 Specimen Preparation

- Semen sample is collected with :
 - **Abstinence period** of **2-7 days**.
 - **Ideal collection** through **masturbation** in sterile container.
 - **Non-spermicidal polyurethane semen collection pouch (Sperm Collect™)** can be used when required.
- Semen sample is allowed to liquefy and then well mixed for performing test.
- Ideally test is to be performed within 30 to 60 min of collection.
- **Sperm-free Seminal Plasma Preparation :**
 - **Note down** semen volume (up to one decimal).
 - **Centrifugation** (with Androspin™) of semen sample (liquefied, well mixed) at **3000 rpm** for **10 - 20 min**.
 - **Aspirate supernatant** to obtain seminal plasma & **leave precipitate**.
 - Can be stored at **-20°C** for **12 months**.

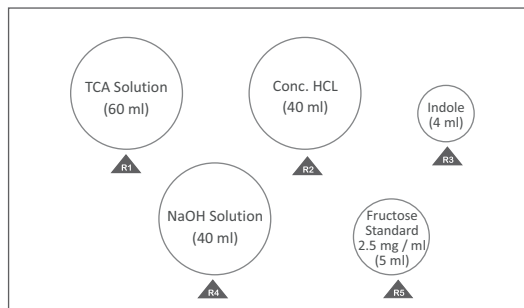
Special Instructions :

- **Hyperviscous** semen sample should be processed to bring towards normal viscosity. (**Viscosity-CH™** or **Viscosity-BR™** kit can be used)
- **Frozen semen** plasma must be thawed at 37°C (with Sperm Warmer™) before performing test.

2 Kit Contents

- Reagent 1 (R1) TCA Solution : 60 ml
- Reagent 2 (R2) Conc. HCL : 40 ml
- Reagent 3 (R3) Indole : 04 ml
- Reagent 4 (R4) NaOH Solution : 40 ml
- Reagent 5 (R5) Fructose Standard (2.5 mg / ml) : 5 ml

Kit Content Layout Diagram :



Storage Conditions :

- The kit should be stored in dark at 2°C - 8°C after receiving.
- Bring all the reagents to room temperature before use.
- Once open store reagents in the fridge protected from light.
- Expiry date is printed on the out side of the box.

REQUIRED BUT NOT PROVIDED IN KIT

- Controlled Temperature 37°C Dry bath (Sperm Warmer™ / Water bath)
- Centrifuge Machine (Androspin™)
- Bio-chemistry Analyser (Androchem Analyser™)
- Pipettes Set
- Stopwatch
- Microtip Box
- Test Tube Stand

REQUIRED BUT NOT PROVIDED IN KIT

- Hand gloves
- Semen Collection Container
- Non-spermicidal Semen Collection Pouch (Sperm Collect™)
- Microtips
- Pasteur Pipettes
- Test Tubes
- Micro Tubes

Assay Parameters For Programming :

- | | |
|-----------------------------|--------------|
| 01. Test Name | : Fructose |
| 02. Mode | : END POINT |
| 03. Primary wavelength | : 510 nm |
| 04. Temperature | : 37°C |
| 05. Aspiration Volume | : 500 µL |
| 06. Lag time | : 05 sec |
| 07. Blank | : Yes |
| 08. Q. C. | : No |
| 09. Standard | : 01 |
| 10. Concentration | : 2.5 mg/mL |
| 11. Normal | : Yes |
| 12. Reagent linearity | : 40 µmol/mL |
| 13. Read sec | : -- |
| 14. K factor | : 5.416 |
| 15. Unit | : µmol/mL |
| 16. Reaction Time | : 60 min |
| 17. Reagent Blank Abs. max. | : 0.5 |

NOTE :

AndroChem[™]
Analyser



(Mfg. by Sperm Processor Pvt. Ltd., Aurangabad, India)

- Is preprogrammed with above mentioned settings.
- No additional programming is required.

Step 1 : • **Note down semen volume** (upto one decimal).

- **Bring** all the reagents to room temperature before use.



Step 2 : Take **two** micro centrifuge tubes & **label** them as '**Standard (Std.) (S)**' & '**Test (T)**'. (With appropriate **Lab ID** No.)



Step 3 : Take the reagents & put in appropriate tubes with help of air displacement pipettes as per the following table.

	Tubes	
	Std.	Test
R1 - TCA Solution	500 μ L	500 μ L
R5 - Fructose Standard	100 μ L	----
Semen / Seminal Plasma	----	100 μ L

Step 4 : Mix well the solutions of **step 3**.

Step 5 : Centrifuge the **step 4** solution tubes at **3000 rpm** for **10 min**.

Step 6 : Remove the supernatants in another test tubes & label them as '**Std. (S1)**' & '**Test (T1)**' respectively.



Step 7 : Take three new test tubes & label them as '**Blank (B)**', '**Std. (S3)**' & '**Test (T3)**'.

Step 8 : Transfer reagents sequentially according to the table given below in '**Blank (B)**', '**Std. (S3)**' & '**Test (T3)**'. Supernatant used are from **Step 6**.



	Tubes		
	Blank	Std. 3	Test 3
R2 - Conc. HCl	250 μ L	250 μ L	250 μ L
Supernatant of Std.	----	25 μ L	----
Supernatant of Test	----	----	25 μ L
R3 - Indole Reagent	25 μ L	25 μ L	25 μ L
<ul style="list-style-type: none"> • Incubate test tubes at 37°C (Dry bath) for 60 min. • After 60 min, add 			
R4 - NaOH Solution	250 μ L	250 μ L	250 μ L

Mix well the solutions of **step 8**.

Step 9 : Tubes [**Blank (B)**, **Std. 3 (S3)** & **Test 3 (T3)**] are now ready for examination.

EXAMINATIONS OF TEST TUBES :

Read the absorbance by using Bio-chemistry analyser (at 510 nm) within 60 min in following order :

Blank	(B)	(Abs. B)
Std.	(S3)	(Abs. S)
Test	(T3)	(Abs. T)

CALCULATION : FRUCTOSE IN SEMEN PER EJACULATE**1. Fructose in semen (Y) (µmol/mL)****A. If Biochemistry Analyser is used :**

- Fructose in semen (**X**) (mg/mL)

$$= \frac{(\text{Abs. T}) - (\text{Abs. B})}{(\text{Abs. S}) - (\text{Abs. B})} \times 2.5 \text{ mg/mL}$$
- Fructose in semen (**Y**) (µmol/mL)
 = Fructose in semen (mg/mL) × 5.416
 = Fructose in semen (µmol/mL)

Note : Correction factor 5.416 is calculated on the basis of sample dilution & incubation time.

- Final Result
 Fructose in semen sample = **Y** µmol / mL

B. If AndroChem Analyser is used :

(Mfg. by Sperm Processor Pvt. Ltd., Aurangabad, India)

- Is preprogrammed with above mentioned settings.
- No additional programming is required.
- Results are displayed as **Y** µmol/mL

2. Fructose in semen (Z) (µmol/ejaculate)

= Fructose in semen **Y** (µmol/mL) × semen volume (mL)

= Fructose in **Z** µmol / ejaculate

Quantitative Estimation of Fructose in Seminal Plasma

(Biomarker - Seminal Vesical)

Volume : ____ mL**Result** : ____ μmol / mL: ____ μmol / ejaculate**Normal Reference Range :** **$\geq 13 \mu\text{mol}$ / ejaculate**

(As per fifth edition of WHO laboratory manual for examination and processing of human semen).

Limitations :

- This test provides presumptive quantitative information of fructose in seminal plasma.
- This parameter should be analyzed by a specialist.
- The result should be evaluated taking into account all clinical & laboratory findings related to the same sample.

- All patient samples & reagents should be treated as potentially infectious & the user must wear protective gloves, eye protection & laboratory coats when performing the test.
- The kit should be discarded in a proper biohazard container after testing.
- Do not eat, drink or smoke in the area where specimens & kit reagents are handled.
- Do not use beyond the expiration date which appears on the package label.
- It is recommended to use of gloves & face mask.

- Do not release the products used into the environment. Follow centre guidelines for the storage & disposal of toxic substances.
- Biological samples must be handled as potentially infectious.

Description of Symbols



consult instructions of use



product reference



lot number



use by



manufacturer



health surveillance device
for in-vitro diagnostic



contains sufficient for 'n' tests



temperature limitation



keep dry



CE mark (Conformité Européene)

Accreditations & Registered Certificates

- **ISO 13485 : 2003** Certified
- **CE** Certified
- **GMDN** Registered
- **US FDA** Registered

For more information & procedure videos



www.spermprocessor.com/sft-fructose.html



www.youtube.com/watch?v=FyoAz54A-n8

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