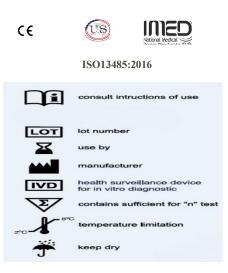
# Ideh Varzan Farda

#### Ideh varzan farda (IVF Co)

Establishment in 2014. IVF Co is one of the leading companies, in research, development and manufacture of sperm analysis reagents and disposable Products for human reproduction .We are trying to produce with same quality as famous brands, but in lower prices.

## Sperm DNA Fragmentation Assay Kit

### SDFA KIT(50 TESTS)



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#### Sperm DNA fragmentation kit

Subfertility affects between 15% - 25% of couples. Approximately 20% of infertile couples, the male partner is the sole cause. Male infertility can be due to a variety of conditions. DNA integrity was recognized as a significant sperm functional parameter with an important role in human ART. That unique chromatin structure protects father's genome from external influences enabling its correct transfer towards oocyte. The DNA integrity is the sperm functional parameter that together with conventional semen parameters gives more reliable and precise diagnosis of male reproductive potential.

#### Precautions

- The test should be discarded in a proper biohazard container after testing.

- Do not eat, drink or smoke in the area where specimens and kit reagents are handled.

- Do not use beyond the expiration date, which appears on the package label.

- The use of gloves and face mask is recommended

. -Do the test under the chemical hood

#### Store condition

After receiving the kit store at 2-8 centigrade for 8 months.

#### Sperm sample

Fresh semen samples should be collected in a sterile container. The sperm DNA fragmentation assay should be performed.

- Staining process, add C Solution on the Incubated for 75 seconds. Then, remove the fixator solution completely. Apply Solution Incubated for 3minutes. Then, remove the stain by tilting and the end adding solution E, incubated for 2 minutes. Remove the excess of stain with water and allow to dry at room temperature.
  - Visualize under bright field microscopy and counting 300 sperms in C and S well.

#### Results

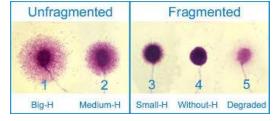
#### **Abnormal Sperms**

- Small halo  $\leq 1/3$  of the minor diameter of the core.

- Spermatozoa without halo.

#### Normal sperms

- Big halo and medium halo  $\ge 1/3$  of the minor diameter of the core.

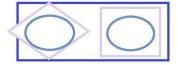


 $SDF(\%) = \frac{Abnormal halo}{total halo counted} \times 100$ 



- NORMAL SDF≤15%
- BORDELINE SDF ~15-30%
  - ABNORMAL SDF≥30% -

Immediately after, transfer 50 ml of the sperm sample to the tube and mix gently with a micropipette. The formation of bubbles shall be prevented. Following, place 30 µl of the cell suspension onto the center of sample wells (S and C). Cover wells with a coverslip. Press gently, avoiding air bubbles formation. Transfer the slide into the fridge at 4°C, for 5 minutes to solidify the agarose.



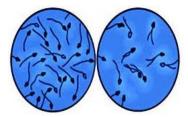
- Take the slide out of the fridge and remove the coverslip slowly. All the Processing must be performed at room temperature (22 °C) and under the chemical hood.
- Place the slide horizontally on staining tray. Apply Solution A on the well making sure the whole well covered with solution and incubate for 7 minutes. Then, remove the reagent without shaking completely.
- Apply Solution B on the well making sure it is fully immersed. Incubate for 15 minutes. Then, remove the reactive by tilting until completing the drying.
- Wash the slide for 5 minutes with abundant distilled water then, Dehydrate by flooding with 70%, 90% and 100% ethanol, incubate for 2 minutes respectively. After drying, processed slides may be kept at room temperature in a dry and dark place for several months.

#### **Description of kit reagents**

- Agarose Cell Support; 50 micro tube
- Pre-treated Slides; 25 units( 50 wells)
- Solution A, Denaturant Agent, 25 cc.
- Solution B, Lysis Solution, 25 cc.
- -Solution C, fixing solution, cc.
- Solution D Staining Solution, 25 cc.
- Solution E Staining Solution, 25 cc.

**Sample preparation** 

• Dilute the sperm sample with washing sperm media or PBS 1X buffer to a maximum of 20 million sperm per milliliter, if the count of sperm is law centrifuge the semen for 5-7 minutes in 1200 rpm and use the sediment.



#### **INSTRUCTION FOR USE**

 Place the agarose tube into the float and melt by using a water bath at 95-100°C for 5 minutes. To melt the agarose can use a microwave. Then keep the tube to be used at 37°C for 1 minute.