



Tosoh Inc.

Reproductive Immunoassays in IVF clinics

In-vitro fertilization (IVF) and intra-uterine insemination (IUI) are the most common methods used by fertility clinics. The overall goal of IVF treatment is to increase the number of follicles, prevent premature ovulation (reduce LH) and create the conditions for successful implantation of a fertilized egg.

After the clinic has evaluated the couple for several male and female fertility factors, in addition to the woman's age, they determine the most appropriate treatment option for the couple. One of the determining factors for egg quality is the Day-3 FSH value. A high FSH predicts poor egg quality. This determines the treatment protocol (GnRH agonist or antagonist protocol).

Once a particular treatment protocol has been determined there are 5 different stages of the IVF procedure.

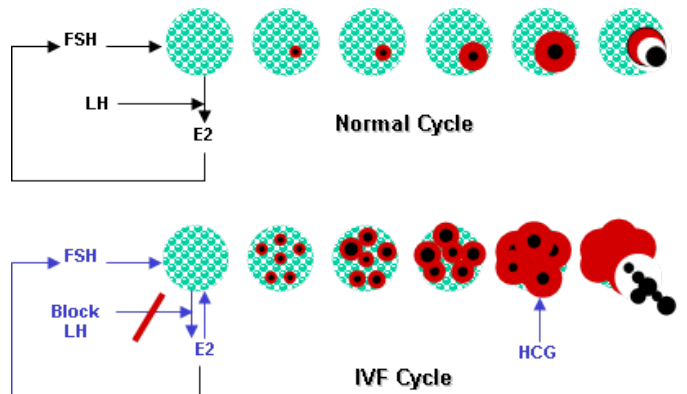
1. Ovarian stimulation: The goal in this stage is to obtain at least 8-10 mature eggs. The stimulation cycle consists of 8 days starting from Day 2 of the menstrual cycle. Blood work and ultrasound are done 3-4 times during this period. The blood work usually consists of E2 and FSH. There is a direct correlation between the E2 value and the number of eggs/follicles. The physician uses the E2 and ultrasound result to determine if the treatment is going well. If a patient is hyper-stimulated and develops too many eggs, the cycle is usually cancelled. It is therefore very important that the physician be able to review the E2 result and ultrasound on the same visit. Tosoh's ST AIA-PACK E2 offers results in 18 to 20 minutes. If the blood is drawn as soon as the patient checks-in, the E2 result will be available to the clinician during the ultrasound.

2. hCG trigger: An hCG injection is given on the 9th or 10th day to induce ovulation and final egg maturation. This depends on the size of a mature follicle. A follicle is considered mature if the diameter is approximately 18-20 mm. The E2 level at this stage is around 1000-4000 pg/mL. On the day of the HCG trigger, the E2/follicle ratio should be approximately 100-200 and the progesterone level should be 0.8 ng/mL. Since E2 levels are very high, it is desirable to have an assay that offers a broad assay range. Tosoh's ST AIA-PACK E2 offers an assay range of 25 to 3000 pg/mL.

3. Egg retrieval: The egg retrieval is planned for 34-35 hours after hCG injection and shortly before that time, ovulation may begin. At this stage the patient should produce enough progesterone for a successful implantation. Progesterone supplements are usually given to prepare the uterus for the implantation.

4. Embryo transfer: After retrieval, the egg is fertilized with the partner's sperm. The embryo can be transferred on day 3 of fertilization or day 5 (blastocyst) of fertilization. The embryo is carefully placed into the patient's uterus. Usually 2-3 embryos are transferred and vary from patient to patient.

5. Beta hCG test: If the implantation is successful, the beta hCG test will be positive on the 14th day after egg retrieval. A positive beta hCG indicates pregnancy. The beta hCG doubles very 48 hours and is monitored throughout the pregnancy. The patient usually begins routine pre-natal care by her obstetrician at 12 weeks of pregnancy.



IUI: In an IUI procedure the patient goes through stages 1 and 2. The stimulation protocol is usually mild to get 1-2 mature follicles. Thirty-four to thirty-five hours after the hCG trigger, the patient is inseminated with the partner's sperm. The fertilization occurs naturally.

Egg Donation: If an egg donor is used, the donor goes through stages 1 to 3. Once the egg is retrieved, it is fertilized with the partner's sperm. The recipient goes through all the stages but the embryo transferred utilizes the egg from the donor.

New AIA-PACK Update
ST AIA-PACK Cystatin C and ST AIA-PACK HbA1c assays have been FDA cleared.
Product will be available by the end of January 2011.

(continued on reverse)

TBI Tech Talk

(continued)

The IVF process is very involved and has an emotional impact on both the patient and the care givers. Because it is very time sensitive, an assay with fast turnaround time is always desirable. Operators using immunoassay analyzers have to multitask, as both embryologist and andrologist in many clinics. A simple, easy to operate instrument is a good choice. The most important criterion for immunoassays in IVF clinics is the reliability of the assays. An E2 assay with low CVs is important as the day-to-day change in result is monitored throughout the stimulation cycle.

Tosoh offers a very good solution with its menu of reproductive assays. The assay time is 18-20 minutes, which allows the physician time to review the results with the patient on the same visit. Tosoh minimizes heterophile antibody interference by using special Ab coating methods and HAMA blockers in the assays. CVs on the E2 are <8.9%. 90 day calibration stability allows the laboratory to test throughout the cycle, with no re-calibration. All of these features of Tosoh's reproductive assays ensure consistent, reliable results.

Assay	AIA-PACK Part #	Sensitivity	Assay Range	Calibration Stability
ST βHCG	025261	0.5 mIU/mL	Up to 400 mIU/mL	90 days
ST FSH	025265	1.0 mIU/mL	Up to 200 mIU/mL	90 days
ST LH II	025296	0.2 mIU/mL	Up to 200 mIU/mL	90 days
ST E2	025274	25 pg/mL	Up to 3000 pg/mL	90 days
ST PROG	025281	0.1 ng/mL	Up to 40 ng/mL	90 days

Q: I recently moved to a Tosoh platform; where do I obtain information on regulatory compliance?

A: It is always advisable to check the latest CLIA guidelines online at <http://www.cms.gov/clia/>. The Clinical Laboratory Improvement Amendments of 1988 (CLIA) established quality standards for all laboratory testing to ensure the accuracy, reliability and timeliness of patient test results regardless of where the test is performed. Useful links are available on topics such as:



Dr. John Murphy
Senior Manager
Technical Support

- Verification of Performance Specifications
- Calibration and Calibration Verification
- Equivalent Quality Control Procedures
- Laboratory Director Responsibilities
- Proficiency Testing

Flag Utility on the G7 and G8 Analyzers

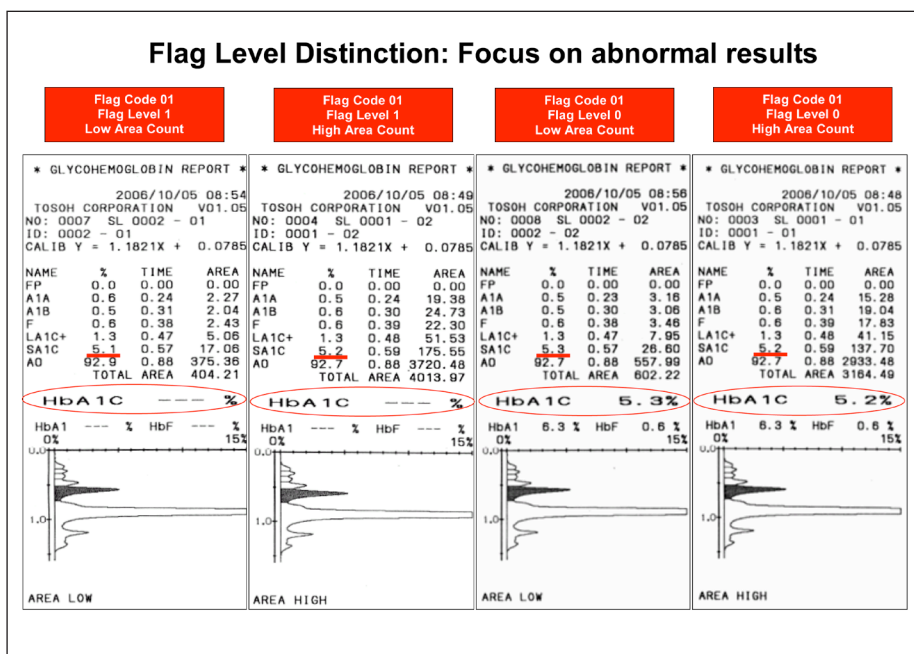
Flags are used to simplify chromatogram review and focus on abnormal results. When a chromatogram is generated, a flag code is triggered and the corresponding flag message is printed on the chromatogram. Flags are not a substitute for chromatogram review, but rather enable the review process. Interpretation must be done with care when results are flagged, and settings may vary depending on the user's choice.

It is possible to program up to 10 flags on the G7 analyzer and 20 flags on the G8 analyzer. Each flag has at least three parts: 1) Flag code (pre-programmed), 2) Flag condition, 3) Flag message (user customized) and 4) Flag level (only available on the G8).

On the G8 instruments, a flag level (0 or 1) can be applied to each of the flag codes:

Flag level 0: The flag is printed together with the result, but interpretation must be done with care.

Flag level 1: The flag is printed but the result is not printed, as this is not valid. Results are printed as "----". This is an additional aid to prevent incorrect results from being transmitted to the host.



Programming flags

To program a flag in the system, please follow these simple instructions:

1. Go to MENU/UTILITY/FLAG.
2. Select an empty field and enter the specific flag code and condition (ex: 01 < 500), which are found in the Operator's and Training Manuals.
3. Enter flag level 0 or 1 (only on G8) and press ENTER.
4. Press MSG EDIT.
5. Enter the flag message (suggested action or action operator must take) and press ENTER.
6. The flag is programmed and will be printed on the chromatogram when a sample is analyzed and meets the flag code criteria.



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