Instructions for use

Anti-Zona Pellucida Antibody Latex Agglutination Test

Cat. No.: BS-20-10

Size: 50 Determinations
Storage: 2 °C – 8 °C (36 °F – 46 °F)

Screening test for the determination of anti-zona pellucida antibodies in serum, cervical mucus, uterine fluid and follicular lavage fluid

- For *in vitro* diagnostic use only -

CE EU Registration No.: DE/CA80/IVD1705

Certified Quality Management System according to
DIN EN ISO 9001:2000
Register No.: IC 373 038, certified by ifta-CERT
DIN EN ISO 13485
Register No.: CE 0483-0215, certified by mdc

BIOSERV DIAGNOSTICS

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Also available from BIOSERV Diagnostics:

Gastroenterology	Order Code	Determinations per Kit
Elastase-1 ELISA (pancreatic insufficiency)	BS - 86 - 01	96
Pankrin™ ELISA (acute pancreatitis)	BS - 86 - 02	96
Infertility		
Sperm Antibody Latex Agglutination Test	BS - 10 - 10	50
Sperm Antibody ELISA	BS - 10 - 20	96
Sperm Antibody Haemagglutination Test	BS - 10 - 30	40
Sperm Antibody ELISA - Ig Typing	BS - 10 - 50	96
Zona Pellucida Antibody Latex Agglutination Test	BS - 20 - 10	50
Zona Pellucida Antibody ELISA	BS - 20 - 20	96
Zona Pellucida Antibody Haemagglutination Test	BS - 20 - 30	40
Zona Pellucida Antibody ELISA - Ig Typing	BS - 20 - 50	96
Ovary Antibody Latex Agglutination Test	BS - 40 - 10	50
Ovary Antibody ELISA	BS - 40 - 20	96
Ovary Antibody Haemagglutination Test	BS - 40 - 30	40
Ovary Antibody ELISA - Ig Typing	BS - 40 - 50	96
Monitoring of Risk Pregnancies		
IGF-BP1 ELISA (PP12)	BS - 30 - 10	96
Glycodelin ELISA (PP14)	BS - 30 - 20	96
Hormones, Tumour Markers		
FSH ELISA	BS - 85 - 21	96
HCG ELISA	BS - 85 - 22	96
LH ELISA	BS - 85 - 23	96
HPL ELISA	BS - 85 - 24	96
Prolactin ELISA	BS - 85 - 25	96
AFP ELISA	BS - 90 - 21	96

Intended Use

The Anti-Zona Pellucida Antibody Latex Agglutination Test from BIOSERV Diagnostics is a quick, reliable, semiquantitative test for the detection of antibodies directed against human zona pellucida. This test may be used with serum, uterine fluid, cervical mucus and follicular lavage fluid.

Clinical Relevance

Antibodies directed against zona pellucida antigens in serum can cause infertility of women.

Fields of Application

The application of the Anti-Zona Pellucida Antibody Latex Agglutination Test from BIOSERV Diagnostics is recommended for monitoring disorders of fertility and premature ovarian failure.

Principles of the Assay Method

In the case of presence of specific antibodies directed against zona pellucida antigens in the sample, latex particles coated with antigen will agglutinate within 1 - 2 minutes.

Reagents

(sufficient for 50 determinations)

1. Zona pellucida antigen suspension (yellow screw cap)	0.55 ml
2. Positive control (green screw cap)	0.3 ml
3. Negative control (red screw cap)	0.3 ml
4. Dilution buffer, concentrated 3x (mix before use with 60 ml distilled water)	30 ml
5. Stirrer sticks	10 x
6. Slides	5 x

Materials Required but not Included

1. Tubes for the dilution of the samples.
2. Distilled or deionised water.
3. Microliter pipettes with disposable tips: 10 µl, 20 µl, 100 µl and 500 µl.
4. Please use only calibrated pipettes.

Warnings and Precautions

1. This kit is intended for *in vitro* use only.
2. Do not pipette reagents by mouth.
3. Please regard all samples as potentially infectious and handle them with utmost care.
4. Handling and disposal should be in accordance with the procedures defined by an appropriate national biohazard safety guideline or regulation where this exists.

Instructions for Reagent Preparation

1. The components of this kit are intended for use as an integral unit and should not be interchanged with the components of other kits.
2. All reagents and specimens must be brought to room temperature before use.
3. All reagents have to be mixed without foaming.
4. Once the test procedure has been started, all steps should be carried out without interruption.
5. Use new disposable tips for each specimen.

Storage Instructions and Shelf Life Information

1. Store the reagents at 2 °C – 8 °C (36 °F – 46 °F).
2. The reagents remain stable until the expiration date of the kit.
3. Put caps back on the vials immediately after use.

Sample Material

Serum, cervical mucus, uterine fluid and follicular lavage fluid

Specimen Collection and Preparation

Serum:

Collect blood by venipuncture, allow to clot, and separate serum by centrifugation at room temperature; avoid haemolysis. Avoid repeated freezing and thawing. Store tubes closed as they may be a danger of contamination or alteration of concentration.

1. Handle all samples with utmost care since they may be infectious.
2. There are no known interferences with extrinsic factors or other substances.
3. Samples may be stored at different temperatures for the following time-spans:
   - Environmental temperature up to 30 °C (86 °F): up to three days
   - Refrigerator temperature (2 – 8 °C / 36 °F – 46 °F): up to one week
   - Household freezer temperature (-10 °C – -20 °C / 14 °F – -4 °F): up to one year

ATTENTION! There are no test methods available which may guarantee that Hepatitis B virus, Human Immunodeficiency Virus (HIV/HTLV-III/LAV), or other infectious agents are absent from the reagents in this kit. Therefore, all human blood products, including patient samples, should be considered potentially infectious.

Assay Procedure for Cervical Mucus, Uterine Fluid and Follicular Lavage Fluid

1. Preparation of dilution buffer: Dilute the concentrated dilution buffer (30 ml) with 60 ml distilled water.
2. Dilute specimen 1 : 50 (10 µl of specimen + 490 µl of dilution buffer).
3. Thoroughly mix specimen and buffer and centrifuge about 10 minutes at 1000 g.
4. Make a serial dilution of supernatant using log 2 (1:100, 1:200, 1:400).
5. Use positive and negative controls undiluted.
6. **Vigorously mix the antigen suspension before use, for example by using a Vortex™mixer for at least 1 minute.**
7. Dispense 10 µl of antigen suspension into the marked sector (circular shape) on the slide. Add 20 µl of controls and diluted specimen (1:100, 1:200, 1:400).
8. Intensively mix the antigen suspension and samples on the slide using one of the stirrer stick provided.
9. Move the slide slowly by hand.
10. Inspect the slide visually for agglutination after 2 min.

Please note that an agglutination is considered to be positive with regard to the presence of anti-zona pellucida antibodies only in specimen dilutions of 1:100 and higher. In case of agglutination please carry on with titration until no more agglutination appears.

Assay Procedure for Serum

1. Preparation of dilution buffer: Dilute the concentrated dilution buffer (30 ml) with 60 ml distilled water.
2. Dilute specimen 1:200 (for example: 5 µl of serum + 995 µl of dilution buffer).
3. Make a serial dilution using log 2 (1:400, 1:800, 1:1600).
4. Positive and negative controls have to be used undiluted.
5. **Vigorously mix the antigen suspension before use, for example by using a Vortex™mixer for at least 1 minute.**
6. Dispense 10 µl of antigen suspension into marked sector (circular shape) on the slide. Add 20 µl of controls and diluted specimen (1:200, 1:400, 1:800, 1:1600).
7. Intensively mix antigen suspension and samples on the slide using a stirrer stick provided.
8. Move slide slowly by hand.
9. Inspect the slide visually for agglutination after 2 min.

Please note that an agglutination is considered to be positive with regard to the presence of anti-zona pellucida antibodies only in specimen dilutions of 1:200 and higher. Therefore in case of agglutination please carry on with titration until no more agglutination appears.

Evaluation of the Results

1. Positive reaction: After 2 minutes a definite agglutination characterised by a more or less coarse granulation indicates a positive result.
   For serum a positive response in the 1:200 dilution is equivalent to 10 IU/ml in the BIOSERV-ELISA (order code BS-20-20).
2. Negative reaction: A result is considered negative if no definite agglutination is discernible. In this case the reaction mixture remains liquid with a milky appearance.

Attention: Please don't wait longer than 3 minutes after start of the agglutination reaction, otherwise the results may become unclear, because of evaporation effects.

Limitations of the Assay

- At temperatures higher than 30 °C (86 °F) the samples should be transported cooled or refrigerated.
- Severely haemolytic or lipaemic sera or sera from patients with liver diseases should not be used. Results may be adversely affected by certain pathologic conditions, such as poly- and monoclonal gammapathies, autoimmune diseases or by an altered immune status.