PartoSure Test

PLACENTAL ALPHA MICROGLOBULIN-1 IMMUNOASSAY KIT
INSTRUCTIONS FOR *IN VITRO* DIAGNOSTIC USE ONLY | STORE IN DRY PLACE AT 15 TO 25°C (59 TO 77°F)

PartoSure™

INTENDED USE

The PartoSure test is a rapid, qualitative test for detecting the presence of placental alpha microglobulin-1 (PAMG-1) in cervicovaginal secretions. The device is indicated as an aid to rapidly assess the risk of spontaneous preterm delivery in ≤ 7 days from the time of cervicovaginal sample collection in pregnant women with signs and symptoms of early preterm labor, intact amniotic membranes and minimal cervical dilatation (< 3 cm), sampled between 24 weeks, 0 days and 34 weeks, 6 days gestation in women with a singleton gestation.

SUMMARY AND EXPLANATION OF THE TEST

An accurate risk assessment of spontaneous preterm delivery ≤ 7 days is clinically important among pregnancies with signs or symptoms suggestive of preterm labor. This is particularly true with respect to both the administration of corticosteroids, which have an optimal benefit ≤ 7 days of administration,[1] as well as the transfer of patients to a tertiary care center capable of caring for the birth of a premature infant. Placental alpha microglobulin-1 (PAMG-1), a protein released from decidual cells into the amniotic cavity throughout pregnancy, was first described by Petrunin et al. in 1975.[2,3] PAMG-1 is not found in the extracellular matrix surrounding the amniotic cavity, which theoretically reduces the opportunity for its unnecessary release into the vaginal cavity due to cervical disturbances caused by digital examinations. The PartoSure test provides an additional option for clinicians to assess the risk of spontaneous preterm delivery within ≤ 7 days in women with a singleton gestation with the benefit of being a rapid, point-of-care test, performed with a simple sample collection procedure that does not require a speculum examination or specialized equipment for sample analysis. Its presence in cervicovaginal discharge when labor and delivery are imminent is likely due to the transudation of the protein through pre-existing pores in the chorioamniotic membranes during uterine contractions and, potentially, degradation of the extracellular matrix of fetal membranes due to an inflammatory process of labor.[4] The clinical utility of a rapid and reliable test result based on a specimen collected non-invasively at the point of care represents an additional option to identify preterm labor that may result in spontaneous preterm delivery ≤ 7 days of testing among women with a singleton gestation.

PRINCIPLE OF THE TEST

PartoSure is a lateral flow, immunochromatographic assay designed to identify the presence of human placental alpha microglobulin-1 (PAMG-1). The test employs monoclonal antibodies sufficiently sensitive to detect 1 ng/ml of PAMG-1. For the analysis, a sample of cervicovaginal discharge collected by vaginal swab is extracted into a solvent. The presence of PAMG-1 antigen is then detected by inserting a lateral flow test strip into the vial. The sample flows from an absorbent pad to a nitrocellulose membrane, passing through a reactive area containing monoclonal anti-PAMG-1 antibodies conjugated to a gold particle. The antigen-antibody complex flows to the test region where it is immobilized by a second anti-PAMG-1 antibody. This event leads to the appearance of the test line. Unbound antigen-antibody complexes continue to flow along the test strip and are immobilized by a second antibody. This leads to the appearance of the internal control line.

REAGENTS AND COMPONENTS

The PartoSure test kit includes the following components: the PartoSure test strip in a foil pouch with desiccant, a sterile flocked vaginal swab, and a plastic vial with solvent solution (0.9% NaCl, 0.05% NaN_3, 0.01% Triton X100). No additional materials are required to perform the PartoSure test.

STORAGE AND STABILITY

Store the PartoSure test kit in a dry place at 15 to 25°C (59 to 77°F). The test should not be frozen.

REF TTDT-1-20-US

For technical assistance please contact:
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Harvard Innovation Launch Lab
114 Western Ave.
Boston, MA 02134, USA
www.partosure.com

PSPI-01-01-US DRAFT Rev A
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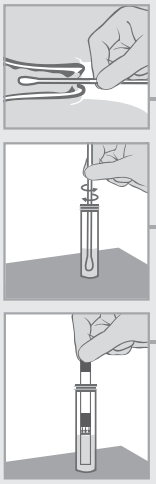
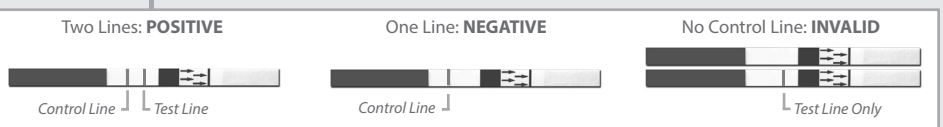
Product of the USA

TEST PROCEDURE



1. Take the solvent vial by its cap and ensure all liquid in the vial has dropped to the bottom. Open the solvent vial and place it in a vertical position.
2. To collect a sample from the vagina, use only the sterile flocked swab provided with the PartoSure test kit. Remove the swab from its package following the instructions on the packaging. The tip of the swab should not touch anything prior to insertion into the vagina. Hold the swab by the middle of its shaft and, while the patient is lying on her back, carefully insert the tip of the swab into the vagina until the fingers contact the skin (no more than 2-3 inches or 5-7 cm deep). Withdraw the swab from the vagina **after 30 seconds**.
3. After the swab has been removed from the vagina, immediately place the tip into the provided solvent vial and rinse by rotating for **30 seconds**.
4. Remove the swab from the vial and dispose of it.
5. Tear open the foil pouch at the tear notches and remove the PartoSure test strip.
6. Insert the white end of the test strip (marked with arrows facing downward) into the vial with solvent.
7. **Remove the test strip from the vial if two lines are clearly visible in the test region or after 5 minutes sharp.** Read the results by placing the test strip on a clean, dry flat surface in a well-lit environment via either natural or flourescent lighting. A positive result is indicated by two lines in the test region, while a negative result is indicated by a single line in the test region. Do not read or interpret the results after 10 minutes have passed since inserting the test strip into the vial.



Positive: Two Red Lines	Negative: One Red Control Line	Invalid: No Red Lines or Test Line Only
Symptomatic patients remain at risk of spontaneous preterm delivery ≤ 7 days	Spontaneous preterm delivery ≤ 7 days is unlikely	Results not valid; collect new sample and retest

The intensity of the lines may vary; the test result is valid even if the lines are faint or uneven. Do not interpret the test result based on the intensity of the lines. Do not read or interpret the results after 10 minutes have passed since inserting the test strip into the vial.

PRECAUTIONS AND WARNINGS

- Use of any swab or solvent solution other than the one provided with the test kit is prohibited.
- Specimens should be collected prior to collection of culture specimens. Collection of vaginal specimens for microbiologic culture frequently requires aggressive collection techniques that may abrade the cervical or vaginal mucosa and may potentially interfere with sample preparation.
- Care must be taken not to contaminate the swab or cervicovaginal secretions with lubricants or antiseptics (e.g. K-Y® or Surgilube® lubricating jelly, Betadine® Cleanser). These substances may interfere with absorption of the specimen by the swab or with the antibody-antigen reaction of the PartoSure test and lead to invalid test results.
- If it is suspected that the patient has applied a topical disinfectant (e.g. Monistat®, miconazole nitrate cream) to the vaginal area within 24 hours, delay specimen collection until 24 hours from application of the topical disinfectant have passed as these products can lead to false negative test results.
- The PartoSure test is not intended for use in women with moderate or gross vaginal bleeding. The presence of vaginal bleeding may contribute to difficulty in interpreting the PartoSure test result. Testing a moderately to grossly bloody sample may lead to false positive results. If upon visual examination you are concerned about the presence of moderate or gross vaginal blood, it is recommended that the sample be collected following the cessation of active vaginal bleeding.
- Specimens should not be obtained from patients with suspected or known placental abruption or placenta previa.
- Do not use the test after the expiration date, which is printed on the product packaging.
- Test kit components are for single-use only.
- Specimens not tested within 24 hours of collection must be stored refrigerated at 2° to 8°C and tested within five days of collection.
- The PartoSure test is for in vitro diagnostic use only and no component of the test should be taken internally.
- Do not use the kit if the swab or test strip package integrity is compromised or if the solvent vial has leaked.
- Do not bend or fold the test strip or the foil pouch with the test strip in it; doing so may damage the strip and lead to inaccurate results.
- The PartoSure test kit includes a plastic vial with solvent solution (0.9% NaCl, 0.05% NaN3, 0.01% Triton X100). Sodium azide may react with plumbing to form potentially explosive metal azides. Avoid contact with skin, eyes, and clothing. In case of contact with any of these reagents, wash area thoroughly with water. If disposing of this reagent, always flush the drain with large volumes of water to prevent azide build-up.
- Safety precautions should be observed when collecting, handling, and disposing of test samples.
- Used test kits are biohazardous; dispose of all components using necessary precautions.

QUALITY CONTROL

The PartoSure test strip contains an internal procedural control mechanism that ensures analytical functionality. The appearance of one or two lines in the results region of the test strip verifies the integrity of the test procedure and components.

LIMITATIONS OF THE TEST

- The PartoSure result should not be interpreted as absolute evidence for the presence or absence of a process that will result in delivery ≤ 7 days from specimen collection.
- The PartoSure test result should always be used in conjunction with information available from the clinical evaluation of the patient and other diagnostic procedures such as cervical examination, assessment of uterine activity, and evaluation of other risk factors.
- The instructions for use must be followed exactly; failure to do so may lead to inaccurate results.
- Test performance has been characterized from specimens taken from the vaginal cavity. Samples obtained from other locations should not be used. A speculum examination is not required.
- Results should be interpreted with caution when a specimen is obtained from a patient with unconfirmed gestational age.
- PartoSure test results are qualitative and not quantitative. No quantitative interpretation should be made based on the strength of the test or control lines.
- This test should only be used in patients with signs and symptoms of preterm labor.
- Invalid test results may occur if lubricants or antiseptics (e.g. K-Y® or Surgilube® lubricating jelly, Betadine® Cleanser) have been used by the patient.
- False positive results may occur if the collection swab is contaminated by moderate to gross vaginal bleeding; false negative results may occur if the collection swab is contaminated by disinfectants (e.g. Monistat®, miconazole nitrate cream).
- While the presence of semen does not interfere with test performance, samples collected immediately following intercourse should be delayed 24 hours if lubricants, antiseptics, or disinfectants are suspected to be present in the vaginal cavity at the time of collection. A speculum examination may be used to reduce the chance of interference by these substances if suspected to be present.
- At this time, information is insufficient to rule out interference by cervical digital examination; therefore, specimens should be collected prior to digital examinations.

EXPECTED VALUES

The PartoSure test detects trace amounts of human PAMG-1. Patient specimens with a concentration of 1 ng/mL placental alpha microglobulin-1 (PAMG-1) or greater will result in a positive PartoSure test. This clinical cutoff was determined using a risk-based approach based on the maximum background concentration of PAMG-1 (0.22 ng/mL) in cervicovaginal secretions to optimize the performance of the PartoSure test. This approach was subsequently validated in a pivotal study including 686 subjects of the intended use population across 15 clinical sites in the United States, and a real-world data study including 511 subjects of the intended use population in one European site.

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SAMPLE COLLECTION AND TEST PROCEDURE

To collect a sample from the vagina, use only the sterile flocked swab provided with the PartoSure test kit. Remove the swab from its package following the instructions on the packaging. The tip of the swab should not touch anything prior to insertion into the vagina. Hold the swab by the middle of its shaft and, while the patient is lying on her back, carefully insert the tip of the swab into the vagina until the fingers contact the skin (no more than 2-3 inches or 5-7 cm deep). A speculum examination is not required. Withdraw the swab from the vagina after 30 seconds. After the swab has been removed from the vagina, immediately place the tip into the provided solvent vial and rinse by rotating for 30 seconds. Remove the swab from the vial and dispose of it. Tear open the foil pouch at the tear notches and remove the PartoSure test strip. Insert the white end of the test strip (marked with arrows facing downward) into the vial with solvent. Remove the test strip from the vial if two lines are clearly visible in the test region or after 5 minutes sharp. Read the results by placing the test strip on a clean, dry flat surface. A positive result is indicated by two lines in the test region, while a negative result is indicated by a single line in the test region. If the control line does not appear (i.e., no lines appear or only the test line appears), then the result is invalid. In the event of an invalid test result, collect a new sample and then retest. Do not read or interpret the results after 10 minutes have passed since inserting the test strip into the vial. The intensity of the lines may vary; the test result is valid even if the lines are faint or uneven. Do not interpret the test result based on the intensity of the lines. Do not read or interpret the results after 10 minutes have passed since inserting the test strip into the vial. Invalid test results may occur if the collection swab is contaminated by lubricants or antiseptics (e.g. K-Y® or Surgilube® lubricating jelly, Betadine® Cleanser).

CROSS REACTIVITY

The PartoSure test was evaluated using a panel of potentially cross-reactive protein substances likely to be found in vaginal specimens including human chorionic gonadotropin, trophoblastic beta-2 glycoprotein, human placental lactogen, alpha-fetoprotein, IGFBP-3, and human serum albumin. Four replicates of each sample containing the potentially cross-reactive substance were tested using a high negative PAMG-1 sample (0.2 ng/mL) and a low positive sample (2.0 ng/mL). Each potentially cross-reactive substance was tested at the highest concentration of substance that was considered clinically relevant. None of the potentially cross-reactive substances tested demonstrated cross-reactivity with the PartoSure test.

INTERFERENCE STUDIES

Lubricants, Disinfectants, Antiseptics, Soaps & Creams

Four (4) replicates each of a high negative sample (0.2 ng/mL) and a low positive sample (2.0 ng/mL) were tested against lubricant jelly, miconazole nitrate disinfectant cream, antiseptic cleanser, body wash soap, and vaginal cream. While all high negative replicates were correctly identified in the presence of miconazole nitrate, low positive replicates were not. The presence of lubricating jelly led to several invalid results for both high negative and low positive replicates, though in cases where a negative or positive result was reported, the result was correct. The presence of antiseptic cleanser led to invalid test results in all replicates. Therefore, care must be taken not to contaminate the swab or cervicovaginal secretions with lubricants or antiseptics (e.g. K-Y® or Surgilube® lubricating jelly, Betadine® Cleanser). If it is suspected that the patient has applied a topical disinfectant (e.g. Monistat®, miconazole nitrate cream) to the vaginal area within 24 hours, delay specimen collection until 24 hours from application have passed as these products can lead to false negative test results. The presence of soap or cream did not interfere with the PartoSure test.

Pharmacological Agents

Four (4) replicates each of a high negative sample (0.2 ng/mL) and a low positive sample (2.0 ng/mL) were tested against 10 non-biologically based interfering substances including: 17-OH progesterone (50 µg/ml), ampicillin (152 µmol/L), cephalexin (337 µmol/L), erythromycin (81.6 µmol/L), gentamycin (21 µmol/L), dexamethasone (1.53 µmol/L), magnesium sulfate (50 µg/mL), oxytocin (58 µU/mL), terbutaline (1 mg/mL), and ritodrine (100 µg/mL). None of these non-biological substances interfered with the PartoSure test.

Vaginal Bacterial Pathogens

Four (4) replicates each of a high negative sample (0.2 ng/mL) and a low positive sample (2.0 ng/mL) were tested against 3 vaginal bacterial pathogens including Gardnerella vaginalis, Candida albicans, and Trichomonas vaginalis. None of these vaginal infection pathogens interfered with the PartoSure test.

Maternal Bleeding

Four (4) replicates each of a high negative sample (0.2 ng/mL) and a low positive sample (2.0 ng/mL) were tested against ten (10) individual maternal blood samples at the three (3) lowest admixture levels determined to represent "trace", "moderate," or "gross" levels of maternal bleeding on the vaginal collection swab. Moderate or gross vaginal bleeding may therefore contribute to difficulty in interpreting the PartoSure test result and may lead to false positive results.

Semen & Urine

A high negative (0.2 ng/mL) sample and a low positive sample (2.0 ng/mL) were tested against 10 individual samples of semen and 10 individual samples of maternal urine. Neither semen nor maternal urine demonstrated interference with the PartoSure test.

PERFORMANCE CHARACTERISTICS

Precision and Reproducibility

Precision and reproducibility were determined using three lots of the PartoSure test at three intended-use sites by three different intended users at each site. Five replicates of seven different PAMG-1 concentrations above and below the limit of detection of the PartoSure test were used, including an absolute zero (0.0 ng/mL), a low negative (0.2 ng/mL), a high negative (0.5 ng/mL), the limit of detection (1.0 ng/mL), a low positive (2.0 ng/mL), and two concentrations within the C_5-C_{95} interval for the device (0.7 ng/mL and 0.9 ng/mL). The results of this study demonstrated 100% negative readings at ≤0.5 ng/mL and 100% positive readings at ≥1ng/mL PMAG-1 levels by intended users.

Safety & Effectiveness

The results of two large clinical studies have been used to demonstrate the safety and effectiveness of the PartoSure test as an aid in assessing the risk of spontaneous preterm delivery ≤ 7 days of testing among women with a singleton gestation and signs and symptoms of preterm labor. A spontaneous preterm delivery was defined as delivery occurring subsequent to spontaneous onset of preterm labor, preterm premature rupture of membranes, or fetal membrane prolapse, regardless of subsequent labor augmentation or cesarean delivery.[5] In the first study (US Study), which was a large, multi-center, prospective cohort study, 686 women with a singleton gestation were evaluated at 15 clinical sites across the United States. From these study data, it was determined that the expression of placental alpha microglobulin-1 (PAMG-1) in vaginal secretions can be used to assess the risk of spontaneous preterm delivery ≤ 7 days of specimen collection among symptomatic pregnant women with a singleton gestation and signs and symptoms of preterm labor. In the second study (European Study), which was a prospectively conducted, retrospective cohort study evaluating the performance of the PartoSure test using real-world data (RWD) from a large maternity hospital in Europe, 511 women with a singleton gestation were evaluated. From these data, it was determined that the findings of the large, prospective cohort study are supported by the real-world performance of the PartoSure test.

In both studies, the PartoSure test specimen was collected prior to cervical digital examination from symptomatic pregnant women with a singleton gestation meeting the following clinical criteria:

- Have signs and symptoms of threatened preterm delivery limited to:
  - o Uterine contractions (with or without pain)
  - o Intermittent lower abdominal pain, dull backache, pelvic pressure
  - o Vaginal bleeding during the second or third trimester
  - o Menstrual-like intestinal cramping (with or without diarrhea)
  - o Change in vaginal discharge (amount, color, or consistency)
  - o Vague sense of discomfort characterized as "not feeling right," which may include such symptoms as general malaise, aches, and/or discomfort that are suggestive of possible preterm labor
- Have a gestational age between 24 weeks, 0 days and 34 weeks, 6 days
- Have intact amniotic membranes
- Have minimal cervical dilatation (< 3 centimeters)

The positive and negative predictive values (PPV/NPV) of the PartoSure test for the prediction of spontaneous preterm delivery ≤ 7 days in both studies are shown in Table 2.

Table 2. PartoSure PPV & NPV for the Prediction of Spontaneous Preterm Delivery ≤ 7 Days Among Symptomatic Women with a Singleton Gestation (n=1,197)

US Study	PartoSure Test	
Statistic	Proportion	% (95% CI)
Positive Predictive Value (PPV)	3/14	21.4 (4.7, 50.8)
Negative Predictive Value (NPV)	669/672	99.6 (98.7, 99.9)
European Study	**PartoSure Test**	
Statistic	Proportion	% (95% CI)
Positive Predictive Value (PPV)	9/29	31.0 (15.3, 50.8)
Negative Predictive Value (NPV)	473/482	98.1 (96.5, 99.1)

For spontaneous preterm delivery ≤ 7 days in women with a singleton gestation, the PPV & NPV of the PartoSure test from the US and European Studies were 21.4% and 99.6%, and 31.0% and 98.1%, respectively. The prevalences of spontaneous preterm delivery ≤ 7 days among women with a singleton gestation in the US and European Studies were 0.9% and 3.5%, respectively.

Additional Performance Characteristics

The sensitivity and specificity of the PartoSure test from the US and European Studies for spontaneous preterm delivery ≤ 7 days were 50.0% (95% CI 11.8, 88.2) and 98.4% (95% CI 97.1, 99.2), and 50.0% (95% CI 26.0, 74.0) and 95.9% (95% CI 93.8, 97.5), respectively. Symptomatic patients with a negative PartoSure test are still at increased risk for spontaneous delivery within 7 days of testing, simply because they present for unscheduled care. It is critical to recognize that PAMG-1 is only one marker of preterm delivery and is not present in elevated levels detectable by PartoSure in 50% of patients who ultimately deliver spontaneously ≤ 7 days of testing. Table 3 provides the 2x2 performance tables in both the US and European studies.

Table 3. PartoSure Test 2x2 Performance Tables (n=1,197)

US Study	sPTD ≤ 7 Days		
PartoSure Test Results	Yes	No	Total
Positive	3	11	14
Negative	3	669	672
Total	6	680	686
European Study	**sPTD ≤ 7 Days**		
PartoSure Test Results	Yes	No	Total
Positive	9	20	29
Negative	9	473	482
Total	18	493	511

In the US study, of the 6 spontaneous preterm deliveries ≤ 7 days, 3 were predicted by the PartoSure test (i.e., true positives) and 3 were missed by the PartoSure test (i.e., false negatives). In the European study, of the 18 spontaneous preterm deliveries ≤ 7 days, 9 were predicted by the PartoSure test (i.e., true positives) and 9 were missed by the PartoSure test (i.e., false negatives).

CLINICAL SIGNIFICANCE OF THE PARTOSURE TEST

Evidence supporting the efficacy of the PartoSure test has been limited to observational studies establishing the association between the test result and likelihood of spontaneous preterm delivery ≤ 7 days. Because no randomized controlled studies have yet been completed to determine the therapeutic efficacy of using the PartoSure test in conjunction with other clinical information for the treatment or prevention of threatened preterm delivery, it is not possible to make recommendations regarding specific treatment options.

Management of Women with a Positive PartoSure Test Result

Management decisions should always be made in conjunction with other available clinical information to avoid unnecessary interventions such as bedrest, in-utero transfers, hospitalizations, and medication administration.

Management of Women with a Negative PartoSure Test Result

While pregnant women with symptoms of preterm labor and a singleton gestation have a pre-test probability as low as 0.9% of spontaneously delivering ≤ 7 days of specimen collection, those with a negative PartoSure test result have as low as a 0.4% probability of delivering spontaneously ≤ 7 days. Despite this low probability, it is important to always consider a negative PartoSure test result in conjunction with all information provided by the patient's clinical evaluation and diagnostic procedure results such as cervical examination, assessment of uterine activity, and evaluation of other risk factors. Patient education regarding her risk of preterm delivery given her symptoms as well as continued vigilance surrounding her condition should remain critical components of her management plan.

ROLE OF PARTOSURE IN IDENTIFYING DELIVERY RISK

The PartoSure test should always be used in conjunction with other clinical risk factors and information to assess overall risk of imminent spontaneous preterm delivery, thus allowing for appropriate patient management. The PartoSure test is an aid in assessing the risk of spontaneous preterm delivery ≤ 7 days from the time of cervicovaginal sample collection in pregnant women with a singleton gestation and signs and symptoms of early preterm labor.

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