- Handle all forensic samples as if they were capable of transmitting disease. Follow standard procedures for proper disposal of specimens.
- Kit reagents contain sodium azide as a preservative which may react with lead or copper in plumbing to form potentially explosive metal azides. Upon disposal, always flush with large volumes of water to prevent build up in drains.

Quality Control

The control line in the control area 'C' can be considered an internal procedural control. A distinct pinkish line will always appear if the test has been performed correctly. If the control line 'C' does not appear, the test is invalid and a new test should be performed following the correct test procedure. A quality control test using positive and negative control standards may also be performed.

Limitations

1. ABAcard® p30 Test is only for in vitro detection of p30 for the forensic identification of semen.
2. The test must be performed in strict accordance with these instructions to obtain accurate and reproducible results.
3. Even if the test result is positive, careful forensic judgment should be made in conjunction with other information and information from other test results.
4. If elevated p30 levels is suspected but a negative result is obtained, the test should be repeated and with fresh specimen.
5. Positive results may be obtained with male urine, which has a reported mean value of 260 ng/ml. Seminal vesicle specific antigen should not be present when tested with urine. Use of another appropriate test is recommended when male urine is in question.
6. Appropriate specimen should be used since p30 is detectable in the vaginal tract only up to a maximum of 2 days.

Performance Characteristics

Sensitivity

The minimum detection limit of ABAcard® p30 Test is 4 ng/ml in 10 minutes (using Stanford's seminal plasma derived Standard, Catalog # L-F500, Phone # (650)725-5542 with PBS /BSA Buffer, pH 7.4, Sigma Catalog # P3688). Results with specimens having high levels of p30 may be obtained as early as 1 minute. For negative results, one must wait for full 10 minutes. The range of p30 is 200,000 to5.5 million nanograms/ml of semen. Therefore, depending on p30 concentration,seminal fluid diluted up to 1 in a million may also be detectable.

Specificity

Hemoglobin (10 g/L), bilirubin (100 mg/L) and lipemic samples, as indicated by triglyceride (5 g/L), do not interferes with the test results. High protein concentration such as prostatic acid phosphatase (1000 ng/ml), albumin (20 g/L), chorionic gonadotropin (900 IU/ml), transferrin (5g/L) and prolactin (1mg/L) did not interfere with test results. Besides semen from both normal and vasectomized men, positive results were obtained from post-ejaculate urine and male urine from adult men, when the urine samples were directly added to the test. However, it is well established that p30 does occur in these urine samples with a reported mean value of 260 ng/ml. Seminal vesicle specific antigen should not be present when this test is used with urine.

Intra Assay and Inter Assay Studies

Intra-assay

An Intra Assay variability study was performed. Ten replicates of known positive and negative p30 samples were tested. The results demonstrated a 100% agreement with the expected results.

Inter-assay

Independent assays were performed on the above samples with three lots of ABAcard® p30 Test over a three month period. The assay results were 100% in agreement with the expected results.

Manufactured by: **Abacus Diagnostics, Inc.**

Some Frequently Asked Questions

Q1. What is "High Dose Hook Effect"?

A1. "High Dose Hook Effect" occurs when the p30 concentration is too high since ABAcard® p30 test is very sensitive. The mechanism behind this effect is that huge amounts of human p30 bind both to the antibody to form an antigen-antibody complex but also free p30 migrates towards the test area "T". The antibody in the test area 'T' is blocked by this free p30. Therefore the mobile antigen-antibody complex with the pink color cannot bind to the antibody. As a result no pink line will form in the test area "T" although a lot of p30 is present in the sample giving a false negative result.

Q2. Is there any minimum and maximum times for reading the results.

A2. Yes there is a maximum time of 10 minutes. The minimum time in a positive result is the time at which both lines appear. The time of the reaction depends upon p30 concentration and other characteristics of the specimen. However if the test line did not appear before ten minutes, one should wait for full 10 minutes to allow the reaction to occur. Specimen with lowest concentration of p30 should take longest time to react. It is to be noted that the results should not be read after 10 minutes since non specific reactions may occur and may result in false positives.

Q3. What does control band 'C' represent?

A3. The built-in procedural positive control is provided by the appearance of a pink line next to the letter 'C', validating the integrity of the test, assuring that the correct test procedure was followed and indicating that proper volume of the fluid entered the test cassette and capillary flow occurred. If the line in the control area 'C' does not develop within 10 minutes, the test result is invalid. Repeat the test using proper procedures.

Q4. Does the intensity of the test band 'T' and control band 'C' matter?

A4. The intensity of either the control band or the test band should not be compared between tests or to each other for ABAcard® p30 Test and no quantitative interpretation should be made based upon differences in the intensity. The mere appearance of both lines proves the presence of p30.

Q5. Where can we order the standard from?

A5. You may order L-F500 standard from Stanford by calling (650) 725-5542.

You may order PBS/BSA buffer from Sigma (Cat # P3688) by calling (800)-325-3010

References

(1) Benton, K.A., Donahue, J.A., Valadez, Jr., M. Analysis of the ABAcard® p30 Test for use in the forensic laboratory. 1998.
(2) Kuester, J., Rothenberg, D., Schwartz, E., Eustace, M., Adamo, R. Validation of a commercial p30 kit (ABAcard®) for forensic identification of semen. 1998.
(3) Carradine, C.C. Evaluation of ABAcard® p30 test for the forensic identification of Semen. 1998.
(4) Kristaly, A., Smith, D.A.S. Validation of ABAcard® p30 test for the rapid forensic identification of Semen, 1999.
(5) Stamey, T. et al. Reference reagents for prostate-specific antigen (PSA): Establishment of the first international standards for free PSA and PSA (90:10). Clinical Chemistry. V 46(9), p 1291-92, 2000.
(6) Cartledge JJ et al. The stability of free and bound prostate-specific antigen.. BJU Int, Nov;84(7):p 810-4, 1999
(7) Sokoll, L.J., Chan, D.W. p30. Its discovery and biochemical characteristics. Urologic Clinics of North America. v 24(2), p253-9, 1997.
(8) Diamandis, E.P., Yu, He. Nonprostatic Sources of Prostate-Specific Antigen. Urologic Clinics of North America. v 24(2), p275-82, 1997.
(9) Stamey, T.A. et al. Identity of p30 purified from seminal fluid by different methods: comparison by amino acid analysis and assigned extinction coefficients. Prostate. v 27(4), p 198-203, 1995.
(10) Jimenez, Verdejo.A., Osuna, E. et al. Study of the enzymatic activity of GGT, LDH, PAP and p30 in semen stains: application to age calculation. For. Sci. Int. v 68(1), p 7-15, 1994.
(11) Armbruster, D.A. p30: biochemistry, analytical methods, and clinical application. Clinical Chemistry. V 39(2), p 181-95, 1993.
(12) Stowell, L.I. et al. An enzyme-linked immunosorbent assay (ELISA) for p30. For. Sci. Int. v 50(1), p 125-38, 1991.
(13) Engelmann, U.H., Schramek, P., Tomanichel, G., Deindl, F., Senge, T.H. Vasectomy reversal in central Europe: results of a questionnaire of urologists in Austria, Germany and Switzerland. J. Urol. v 143(1), p 64-67, 1990.
(14) Graves, H.C.B. et al. Postcoital detection of a male-specific semen protein. Application to the investigation of rape. New. Engl. J. Med. v 312 (6), p 338-343, 1985.
(15) Willot, G.M. Frequency of azoospermia. For.Sci.Int. v 20(1), p 9-10, 1982.
(16) Sensabaugh, G.F.. Isolation and characterization of a semen-specific protein from human seminal plasma: a potential new marker for semen identification. J. Forensic.. Soi. v 23 (1), p 106-115, 1978.