COMPARATIVE STUDIES OF NORMAL AND INFERTILE BOAR SEMEN

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A THESIS SUBMITTED IN PARTIAL FULFILLMENT OF THE REQUIREMENT FOR THE DEGREE OF MASTER OF SCIENCE (BIOCHEMISTRY)

IN THE FACULTY OF GRADUATE STUDIES OF MAHIDOL UNIVERSITY 1986
ABSTRACT

Semen from normal and infertile boar were examine and divided into five groups: normozoospermia, immotile spermatozoa, oligozoospermia, teratozoospermia, and azoospermia according to density, motility and morphology of spermatozoa.

In this study, lectins were used to analyze variations in the distribution of exposed saccharides of the sperm plasma membrane of epididymal spermatozoa, ejaculated normal and abnormal spermatozoa. Three times washed spermatozoa were labeled with lectins conjugated to fluorescein isothiocyanate. The spermatozoa from caput, corpus and cauda epididymis were labeled with FITC-Con A in order to visualize changes in the distribution of exposed membrane glycoproteins during epididymal transit. The cauda epididymal and the ejaculated spermatozoa from normal and infertile boar semen were labeled with FITC-Con A, FITC-PNA, FITC-WGA and FITC-RCA$_{120}$. Con-A binding showed minimal changes during epididymal transit with an increased binding to the principal segment of acrosome after ejaculation. PNA binding was limited to the principal segment of acrosome of cauda epididymal spermatozoa. Strong fluorescence was also found over the principal segment of acrosome of ejaculated spermatozoa but weak fluorescence was also found on the other part of spermatozoa. Con A and PNA binding patterns were not different between normal and abnormal spermatozoa within each lectin used. WGA showed distinct changes in binding patterns on cauda epididymal, ejaculated normal and abnormal spermatozoa. RCA$_{120}$ binding decreased after
ejaculation, however, it showed stronger fluorescence on abnormal spermatozoa than the normal. The different binding patterns on normal and abnormal spermatozoa reflect the different distribution of saccharide groups.

The spermatozoa were solubilized in sample buffer containing 3% SDS, 5% MSH and 10% glycerol then the proteins were analyzed by SDS-PAGE. The 27 K protein was found in immotile spermatozoa whereas it was not found in normal spermatozoa. This protein was shown in 2-dimesional gel to have pI 5.0. The protein patterns of seminal plasma from normal and infertile semen were analyzed by SDS-PAGE. The protein bands of 14-19K were the most abundance in boar seminal plasma. The 96K protein was absent in seminal plasma from oligozoospermia, teratozoospermia and azoospermia. The 130K protein was present only trace amount in azoospermia. All of those proteins may be used as a marker for infertility.

It was also found that the boar seminal plasma proteins contained intermolecular and intramolecular disulfide bonds. The protein band of approx. 105K in teratozoospermia was the reducing product of 130K, whereas the band 130K in all other cases was the reducing product of approx. 105K.

Electrophoretic protein patterns of spermatozoa and seminal plasma proteins were electrophoretically blotted to nitrocellulose sheets and then stained with FITC-lectins. The general similarity in lectin staining reflects the similar seminal plasma glycoproteins from normal and infertile boar semen.
Bound sialic acids in seminal plasma were assayed according to the methods of Massamiri et al. (1979) and Toowicharanont (1983). The contents of the bound sialic acids was not significantly different between normal and infertile boar seminal plasma.