ISOLATION OF NUCLEAR PROTEIN MATRIX FROM HUMAN SPERM HEAD

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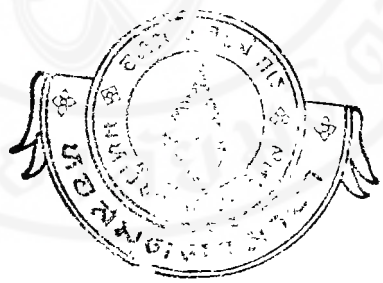

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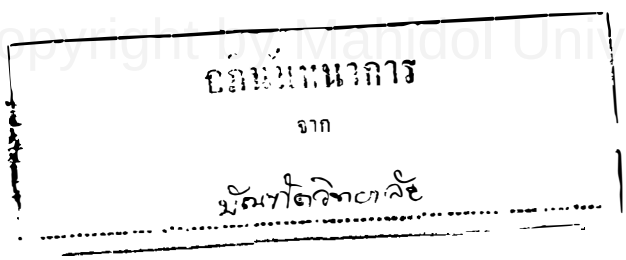

ABSTRACT

The nuclear protein matrix is a residual nuclear structure that can retain its shape after extraction of the major components, basic protein and DNA. This residual structure has been isolated and characterized in many kinds of mammalian somatic nuclei and is thought to play a major role in determining nuclear shape. The purpose of this study was to carry out biochemical and morphological investigations to see whether the nuclear protein matrix exists in the human sperm nucleus.

The isolation procedures involved the removal of nuclear material without destroying the shape of human sperm heads by using different reagents which consist mainly of thiol reagent : DTT or ME, salt : NaCl or $CaCl_2$ - $MgCl_2$ and strong denaturant : urea or Gu-HCl and different mechanical techniques : shearing or agitating. Biochemical analysis was performed in parallel to observations of the integrity of human sperm heads under the electron microscope.

The data obtained from these studies all confirm the existence of a nuclear protein matrix in human sperm nuclei. Extraction with urea, ME, NaCl and DNase I leaves a thin nuclear exoskeleton that can retain the sperm head shape while extraction with Ca^{2+} - Mg^{2+}, DTT and DNase I results in a residual structure appearing as a hollow structure bounded by rather thick ridges or nuclear protein matrix. Acid-urea gel electrophoresis demonstrated the absence of protamines in nuclear exoskeleton but small amounts of them in nuclear protein matrix. SDS-PAGE of both residual nuclear structures show similar polypeptide band pattern with intense bands in molecular weight range of 17,000 - 20,000 and minor bands at 30,000 - 76,000. Amino acid analysis also shows clear similarities among the pellets obtained from different procedures and rat liver nuclear protein matrix.