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Cloning and Sequencing of cDNA Encoding for the Testis-Specific Fox (*Vulpes Vulpes*) Sperm Polypeptide Vb of the Cytochrome C Oxidase

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Identification of fox (Vulpes vulpes) sperm antigens was carried out to assess their interest as a potential target for a contraceptive vaccine. We report here the cloning and sequencing of fSP8, a fox sperm protein of 14.7 kd. fSP8 was isoantigenic in foxes, as it was recognized by sera of both male and female foxes immunized with fox sperm proteins. No glycosylation was detected, on fSP8, as shown both by deglycosylation assay and lectin labeling. To determine the fSP8 sequence, the NH_2-terminal sequence was first obtained, and a piece of cDNA was amplified from testicular RNA by Rapid Amplification of cDNA extremities polymerase chain reaction. This piece was used to screen a cDNA library from fox testis by Southern blot. A sequence of 879 base pairs was obtained, which includes a major open reading frame coding for 128 amino acids. Mass spectrometric analyses have confirmed the position of the open reading frame. Analysis of the predicted amino acids sequence revealed no apparent transmembrane regions. Comparison of the protein sequence with the Prosite database demonstrated a homology with the Zinc binding site of the subunit Vb of the cytochrome c oxidase. On the C-terminal extremity, fSP8 presents a high homology to the Vb polypeptide of the cytochrome c oxidase from bovine, mouse, and human; however the 34 amino acids on the NH_2-extremity were specific to fSP8. Moreover, it was demonstrated that this sequence was testis-specific. This could contribute to the antigenicity of this protein. fSP8 is one of the first fox sperm antigens to be cloned and sequenced.

Key words: Sperm antigen, fox, tissue specificity, immunocontraception

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