3-3-15. Male Specific Protein (MSP) from Wax Moth, *Galleria mellonella* L. ; Purification, Cloning and Characterization

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Male specific protein (MSP) is a soluble protein which is accumulated in high amounts in the hemolymph and other organs of adult male wax moth. The MSP was purified from adult male wax moth by gel filtration and reversed phase column chromatography. This method was slightly modified from the old strategy used in our previous studies. Several internal amino acid sequences of MSP were obtained by in-gel digestion method using trypsin because of its blocked N-terminus.

RT-PCR was conducted using degenerate primers designed from the internal amino acid sequences. 5'-RACE PCR was used to obtain complete protein coding sequence and 5'-UTR. The full length MSP cDNA sequence encodes a 239 amino acid polypeptide including a potential signal peptide region consisting of 18 amino acids. It was theoretically calculated that the recombinant MSP has a molecular mass of 24,317 Da and an isoelectric point (pI) of 6.00 without signal peptide region, but the recombinant protein showed a molecular mass of about 27 kDa similar to that of innate MSP by SDS-PAGE.

Sequence alignment showed the similarity between MSP and juvenile hormone binding proteins (JHBPs) of several lepidopteran species including *G. mellonella*.