

## A : Recombinant myeloperoxidase :

Agilent Spectrum Mill

1	TPQPSEGAAP	AVLGEVDTSL	VLSSMEEAKQ	LVDKAYKERR	ESIKQRLRSG	SASPMELLSY	FKQPVAATR	AVR <b>AADYLHV</b>	80
81	<b>ALDLLER</b> KLR	SLWR <b>RPFNVT</b>	<b>DVLT</b> PAQLNV	<b>LSK</b> SSGCAYQ	DVGVT <b>C</b> PEQD	<b>KYR</b> TITGM <b>CN</b>	<b>NRR</b> SPTLGAS	<b>NRA</b> FVRLPA	160
161	EYEDGFSLPY	GWTPGV <b>K</b> RNG	FPVALAR <b>AVS</b>	NEIV <b>R</b> FPTDQ	LTPD <b>Q</b> ERSLM	FMQWGQLLDH	DLDF <b>T</b> PEPAA	<b>RAS</b> F <b>V</b> T <b>GV</b> <b>N</b> C	240
241	<b>ETS</b> <b>CV</b> Q <b>Q</b> PP <b>C</b>	<b>FPL</b> K <b>I</b> PPNDP	<b>RI</b> KNQAD <b>C</b> IP	<b>FFR</b> SC <b>P</b> AC <b>P</b> G	<b>SNT</b> IR <b>N</b> QIN	ALTSFVDASM	VYGSEEP <b>L</b> AR	<b>NLR</b> <b>N</b> MS <b>N</b> QL <b>G</b>	320
321	<b>LLAV</b> <b>N</b> Q <b>R</b> FQD	<b>NGR</b> <b>ALL</b> PF <b>D</b> N	<b>LHDD</b> <b>P</b> CL <b>L</b> T <b>N</b>	<b>RSAR</b> IP <b>C</b> FLA	GD <b>T</b> RSSEMPE	L <b>T</b> SMHT <b>L</b> L <b>L</b> R	E <b>H</b> NRLATE <b>L</b> K	<b>SLN</b> PRWDGER	400
401	LYQE <b>A</b> R <b>K</b> IVG	<b>AMV</b> Q <b>I</b> IT <b>Y</b> RD	YLPLVLG <b>P</b> TA	MR <b>K</b> YLPT <b>Y</b> RS	<b>YND</b> S <b>V</b> DP <b>R</b> IA	NVFTNAFRYG	HTLIQ <b>P</b> FM <b>F</b> R	<b>LDN</b> RYQ <b>P</b> M <b>E</b> P	480
481	NPRV <b>L</b> SR <b>V</b> F	<b>FAS</b> WR <b>V</b> V <b>L</b> EG	<b>GID</b> P <b>I</b> L <b>R</b> GLM	ATPA <b>K</b> L <b>N</b> R <b>Q</b> N	<b>QIA</b> VDEIR <b>E</b> R	LFEQ <b>V</b> M <b>R</b> IGL	D <b>L</b> PAL <b>N</b> M <b>Q</b> RS	<b>RDH</b> GLPGYNA	560
561	<b>WRR</b> <b>F</b> C <b>G</b> LP <b>Q</b> P	<b>ETV</b> Q <b>L</b> GT <b>V</b> L	<b>RNL</b> K <b>L</b> ARK <b>L</b> M	EQYGT <b>P</b> NNID	I <b>W</b> MG <b>G</b> VSEPL	<b>KRK</b> GR <b>V</b> GP <b>L</b> L	<b>ACI</b> IG <b>T</b> Q <b>F</b> R <b>K</b>	<b>LRD</b> GD <b>R</b> FWWE	640
641	NEGV <b>F</b> SM <b>Q</b> QR	<b>QAL</b> A <b>Q</b> IS <b>L</b> PR	<b>IIC</b> D <b>N</b> T <b>G</b> IT <b>T</b>	<b>VSK</b> NNIFMSN	SYPR <b>D</b> F <b>V</b> NC <b>S</b>	<b>TLP</b> AL <b>N</b> LA <b>S</b> W	<b>REAS</b>	704	

The matched peptides cover 30% (216/704 AA's) of the protein.

Protein Name: **MPO**

Species: **HUMAN**

**PN\_MPO\_01**.user Accession #: 1

MS Digest Index #: 1

Masses are:

pI of Protein: **9.26**

Protein MW: **79635.8** Da

Amino Acid Composition: **A51 C15 D35 E34 F29 G40 H7 I29 K21 L77 M20 N41 P53 Q34 R65 S45 T37 V41 W11 Y19**

MS Digest - Agilent Spectrum Mill Rev. 3.3.078

## B : Recombinant myeloperoxidase incubated in the absence of enzymes:

Agilent Spectrum Mill

1	TPQPSEGAAP	AVLGEVDTSL	VLSSMEEAKQ	LVDKAYKERR	ESIKQRLRSG	SASPMELLSY	FKQPVAATR	AVR <b>AADYLHV</b>	80
81	<b>ALDLLER</b> KLR	SLWR <b>RPFNVT</b>	<b>DVLT</b> PAQLNV	<b>LSK</b> SSGCAYQ	DVGVT <b>C</b> PEQD	<b>KYR</b> TITGM <b>CN</b>	<b>NRR</b> SPTLGAS	<b>NRA</b> FVRLPA	160
161	EYEDGFSLPY	GWTPGV <b>K</b> RNG	FPVALAR <b>AVS</b>	NEIV <b>R</b> FPTDQ	LTPD <b>Q</b> ERSLM	FMQWGQLLDH	DLDF <b>T</b> PEPAA	<b>RAS</b> F <b>V</b> T <b>GV</b> <b>N</b> C	240
241	<b>ETS</b> <b>CV</b> Q <b>Q</b> PP <b>C</b>	<b>FPL</b> K <b>I</b> PPNDP	<b>RI</b> KNQAD <b>C</b> IP	<b>FFR</b> SC <b>P</b> AC <b>P</b> G	<b>SNT</b> IR <b>N</b> QIN	ALTSFVDASM	VYGSEEP <b>L</b> AR	<b>NLR</b> <b>N</b> MS <b>N</b> QL <b>G</b>	320
321	<b>LLAV</b> <b>N</b> Q <b>R</b> FQD	<b>NGR</b> <b>ALL</b> PF <b>D</b> N	<b>LHDD</b> <b>P</b> CL <b>L</b> T <b>N</b>	<b>RSAR</b> IP <b>C</b> FLA	GD <b>T</b> RSSEMPE	L <b>T</b> SMHT <b>L</b> L <b>L</b> R	E <b>H</b> NRLATE <b>L</b> K	<b>SLN</b> PRWDGER	400
401	LYQE <b>A</b> R <b>K</b> IVG	<b>AMV</b> Q <b>I</b> IT <b>Y</b> RD	YLPLVLG <b>P</b> TA	MR <b>K</b> YLPT <b>Y</b> RS	<b>YND</b> S <b>V</b> DP <b>R</b> IA	NVFTNAFRYG	HTLIQ <b>P</b> FM <b>F</b> R	<b>LDN</b> RYQ <b>P</b> M <b>E</b> P	480
481	NPRV <b>L</b> SR <b>V</b> F	<b>FAS</b> WR <b>V</b> V <b>L</b> EG	<b>GID</b> P <b>I</b> L <b>R</b> GLM	ATPA <b>K</b> L <b>N</b> R <b>Q</b> N	<b>QIA</b> VDEIR <b>E</b> R	LFEQ <b>V</b> M <b>R</b> IGL	D <b>L</b> PAL <b>N</b> M <b>Q</b> RS	<b>RDH</b> GLPGYNA	560
561	<b>WRR</b> <b>F</b> C <b>G</b> LP <b>Q</b> P	<b>ETV</b> Q <b>L</b> GT <b>V</b> L	<b>RNL</b> K <b>L</b> ARK <b>L</b> M	EQYGT <b>P</b> NNID	I <b>W</b> MG <b>G</b> VSEPL	<b>KRK</b> GR <b>V</b> GP <b>L</b> L	<b>ACI</b> IG <b>T</b> Q <b>F</b> R <b>K</b>	<b>LRD</b> GD <b>R</b> FWWE	640
641	NEGV <b>F</b> SM <b>Q</b> QR	<b>QAL</b> A <b>Q</b> IS <b>L</b> PR	<b>IIC</b> D <b>N</b> T <b>G</b> IT <b>T</b>	<b>VSK</b> NNIFMSN	SYPR <b>D</b> F <b>V</b> NC <b>S</b>	<b>TLP</b> AL <b>N</b> LA <b>S</b> W	<b>REAS</b>	704	

The matched peptides cover 34% (246/704 AA's) of the protein.

Protein Name: **MPO**

Species: **HUMAN**

**PN\_MPO\_01**.user Accession #: 1

MS Digest Index #: 1

Masses are:

pI of Protein: **9.26**

Protein MW: **79635.8** Da

Amino Acid Composition: **A51 C15 D35 E34 F29 G40 H7 I29 K21 L77 M20 N41 P53 Q34 R65 S45 T37 V41 W11 Y19**

MS Digest - Agilent Spectrum Mill Rev. 3.3.078

## C : Recombinant myeloperoxidase incubated in the presence of enzymes

Agilent Spectrum Mill

1	TPQPSEGAAP	AVLGEVDTSL	VLSSMEEAKQ	LVLDKAYKER	ESIKQRLRSG	SASPMELLSY	FKQPVAATR	AVRAADYLHV	80
81	ALDLLERKLR	SLWR <b>RPFNVT</b>	<b>DVLT</b> PAQLNV	<b>LSK</b> SSGCAYQ	DVGVT <b>C</b> PEQD	<b>KYR</b> TITGM <b>CN</b>	<b>NRR</b> SPTLGAS	<b>NRA</b> FVRLPA	160
161	EYEDGFSLPY	GWTPGVK <b>RNG</b>	FPVALARAVS	NEIVR <b>F</b> PTDQ	LTPDQ <b>E</b> RLM	FMQWGQLLDH	DLDF <b>T</b> PEPAA	<b>RAS</b> FVTGV <b>NC</b>	240
241	ET <b>SCV</b> QPP <b>C</b>	FPL <b>K</b> I <b>P</b> NDP	<b>R</b> IK <b>NQ</b> AD <b>C</b> IP	<b>F</b> FR <b>SC</b> PAC <b>PG</b>	<b>S</b> NT <b>IR</b> NQIN	ALTSFVDASM	VYGSE <b>E</b> PLAR	<b>NLR</b> <b>NMS</b> NQLG	320
321	LLAV <b>NQR</b> FQD	NGR <b>ALL</b> PF <b>DN</b>	LHDD <b>P</b> CL <b>L</b> T <b>N</b>	<b>RS</b> AR <b>I</b> P <b>C</b> FLA	GD <b>TR</b> SSEMPE	L <b>TS</b> MHTLLR	E <b>HN</b> RLATE <b>LK</b>	<b>SLN</b> PRWDGER	400
401	LYQ <b>E</b> AR <b>K</b> IVG	AMVQ <b>I</b> ITY <b>RD</b>	YLPLV <b>L</b> G <b>P</b> TA	MR <b>K</b> YLPT <b>Y</b> RS	<b>Y</b> ND <b>S</b> VDP <b>R</b> IA	NVFT <b>NA</b> FRYG	HT <b>L</b> IQ <b>P</b> FM <b>FR</b>	<b>LDN</b> RYQ <b>M</b> EP	480
481	NPRV <b>PL</b> SR <b>V</b> F	FAS <b>WR</b> V <b>V</b> LE <b>G</b>	<b>G</b> ID <b>P</b> IL <b>R</b> GLM	AT <b>PA</b> K <b>L</b> NR <b>Q</b> N	<b>Q</b> IA <b>V</b> DE <b>I</b> ER <b>R</b>	<b>L</b> FE <b>Q</b> VM <b>R</b> IG <b>L</b>	<b>D</b> L <b>P</b> AL <b>N</b> M <b>Q</b> RS	<b>R</b> DHGL <b>P</b> G <b>Y</b> NA	560
561	<b>WRR</b> FC <b>G</b> LP <b>Q</b> P	ETV <b>G</b> Q <b>L</b> GT <b>V</b> L	<b>R</b> N <b>L</b> K <b>L</b> AR <b>K</b> L <b>M</b>	E <b>Q</b> Y <b>G</b> TP <b>N</b> NI <b>D</b>	I <b>W</b> M <b>G</b> V <b>S</b> E <b>P</b> L	<b>K</b> R <b>K</b> G <b>R</b> V <b>G</b> PL <b>L</b>	<b>A</b> C <b>I</b> I <b>G</b> T <b>Q</b> FR <b>K</b>	<b>L</b> R <b>D</b> G <b>D</b> R <b>F</b> W <b>E</b>	640
641	NE <b>G</b> V <b>F</b> SM <b>Q</b> QR	<b>Q</b> AL <b>A</b> Q <b>I</b> SL <b>P</b> R	<b>I</b> IC <b>D</b> NT <b>G</b> IT <b>T</b>	<b>V</b> SK <b>N</b> I <b>F</b> MS <b>N</b>	<b>S</b> Y <b>P</b> R <b>D</b> V <b>N</b> C <b>S</b>	<b>T</b> LPAL <b>N</b> LA <b>S</b> W	<b>R</b> E <b>A</b> S	704	

The matched peptides cover 19% (136/704 AA's) of the protein.

Protein Name: MPO

Species: HUMAN

PN\_MPO\_01.user Accession #: 1

MS Digest Index #: 1

Masses are:

pI of Protein: 9.26

Protein MW: 79635.8 Da

Amino Acid Composition: A51 C15 D35 E34 F29 G40 H7 I29 K21 L77 M20 N41 P53 Q34 R65 S45 T37 V41 W11 Y19

MS Digest - Agilent Spectrum Mill Rev. 3.3.078

## D: Recombinant mutant myeloperoxidase (Q257→T)

1	MGVFFSSSLR	<b>C</b> MVDL <b>G</b> PC <b>WA</b>	GGLT <b>A</b> EM <b>K</b> LL	LALAG <b>L</b> L <b>A</b> IL	AT <b>P</b> Q <b>P</b> SE <b>G</b> AA	PAVLGEVDTS	LVLSS <b>M</b> E <b>E</b> AK	QLVD <b>K</b> AY <b>K</b> ER	80
81	<b>R</b> ES <b>I</b> K <b>Q</b> RL <b>R</b> GS	SAS <b>P</b> M <b>E</b> LL <b>S</b>	Y <b>F</b> K <b>Q</b> P <b>V</b> A <b>A</b> T <b>R</b>	TAV <b>R</b> A <b>A</b> D <b>Y</b> LH	VALD <b>L</b> L <b>E</b> R <b>K</b> L	<b>R</b> SL <b>W</b> R <b>RP</b> FN <b>V</b>	<b>T</b> D <b>V</b> L <b>T</b> PA <b>Q</b> L <b>N</b>	<b>V</b> LS <b>K</b> SS <b>G</b> C <b>A</b> Y	160
161	Q <b>D</b> V <b>G</b> V <b>T</b> C <b>P</b> EQ	D <b>K</b> Y <b>R</b> T <b>I</b> T <b>G</b> M <b>C</b>	<b>N</b> N <b>R</b> S <b>P</b> T <b>L</b> G <b>A</b>	<b>S</b> N <b>R</b> A <b>F</b> V <b>E</b> W <b>L</b> P	A <b>E</b> Y <b>E</b> D <b>G</b> F <b>S</b> L <b>P</b>	Y <b>G</b> W <b>T</b> P <b>G</b> V <b>K</b> R <b>N</b>	<b>G</b> F <b>F</b> V <b>A</b> L <b>A</b> R <b>A</b> V	<b>S</b> N <b>E</b> I <b>V</b> R <b>F</b> P <b>T</b> D	240
241	Q <b>L</b> T <b>P</b> D <b>Q</b> E <b>R</b> S <b>L</b>	M <b>F</b> M <b>Q</b> W <b>G</b> T <b>L</b> L <b>D</b>	H <b>D</b> L <b>D</b> F <b>T</b> P <b>E</b> P <b>A</b>	<b>A</b> R <b>A</b> S <b>F</b> V <b>T</b> G <b>V</b> N	<b>C</b> E <b>T</b> S <b>C</b> V <b>Q</b> Q <b>P</b> P	<b>C</b> F <b>P</b> L <b>K</b> I <b>P</b> P <b>N</b> D	<b>P</b> R <b>I</b> K <b>N</b> Q <b>A</b> D <b>C</b> I	<b>P</b> F <b>F</b> R <b>S</b> C <b>P</b> A <b>C</b> P	320
321	<b>G</b> S <b>N</b> T <b>I</b> R <b>N</b> Q <b>I</b>	N <b>A</b> L <b>T</b> S <b>F</b> V <b>D</b> A <b>S</b>	M <b>V</b> Y <b>G</b> S <b>E</b> B <b>P</b> L <b>A</b>	<b>R</b> N <b>L</b> R <b>N</b> M <b>S</b> N <b>Q</b> L	<b>G</b> L <b>L</b> A <b>V</b> N <b>Q</b> R <b>F</b> Q	<b>D</b> N <b>G</b> R <b>A</b> L <b>L</b> P <b>F</b> D	<b>N</b> L <b>H</b> D <b>D</b> P <b>C</b> L <b>L</b> T	<b>N</b> R <b>S</b> A <b>R</b> I <b>P</b> C <b>F</b> L	400
401	A <b>G</b> D <b>T</b> R <b>S</b> E <b>M</b> P	E <b>L</b> T <b>S</b> M <b>H</b> T <b>L</b> L <b>L</b>	<b>R</b> E <b>H</b> N <b>E</b> L <b>A</b> T <b>E</b> L	<b>K</b> S <b>L</b> N <b>P</b> R <b>W</b> D <b>G</b> E	<b>R</b> L <b>Y</b> Q <b>E</b> A <b>R</b> K <b>I</b> V	<b>G</b> A <b>M</b> V <b>Q</b> I <b>I</b> T <b>Y</b> R	D <b>Y</b> L <b>P</b> L <b>V</b> L <b>G</b> P <b>T</b>	<b>A</b> M <b>R</b> K <b>Y</b> L <b>P</b> T <b>Y</b> R	480
481	<b>S</b> Y <b>N</b> D <b>S</b> V <b>D</b> P <b>R</b> I	A <b>N</b> V <b>F</b> T <b>N</b> A <b>F</b> R <b>Y</b>	G <b>H</b> T <b>L</b> I <b>Q</b> P <b>F</b> M <b>F</b>	<b>R</b> L <b>D</b> N <b>E</b> Y <b>Q</b> P <b>M</b> E	<b>P</b> N <b>P</b> R <b>V</b> P <b>L</b> S <b>E</b> V	<b>F</b> F <b>A</b> S <b>W</b> R <b>V</b> V <b>L</b> E	<b>G</b> G <b>I</b> D <b>P</b> I <b>L</b> R <b>G</b> L	<b>M</b> A <b>T</b> P <b>A</b> K <b>L</b> N <b>R</b> Q	560
561	N <b>Q</b> I <b>A</b> V <b>D</b> E <b>I</b> R <b>E</b>	<b>R</b> L <b>F</b> E <b>Q</b> V <b>M</b> R <b>I</b> G	<b>L</b> D <b>L</b> P <b>A</b> L <b>N</b> M <b>Q</b> R	<b>S</b> R <b>D</b> H <b>G</b> L <b>P</b> G <b>Y</b> N	<b>A</b> W <b>R</b> R <b>F</b> C <b>G</b> L <b>P</b> Q	<b>P</b> E <b>T</b> V <b>G</b> Q <b>L</b> G <b>T</b> V	<b>L</b> R <b>N</b> L <b>K</b> L <b>A</b> R <b>K</b> L	<b>M</b> E <b>Q</b> Y <b>G</b> T <b>P</b> N <b>N</b> I	640
641	D <b>I</b> W <b>M</b> G <b>V</b> S <b>E</b> P	<b>L</b> K <b>R</b> K <b>G</b> E <b>V</b> G <b>P</b> L	<b>L</b> A <b>C</b> I <b>I</b> G <b>T</b> Q <b>F</b> R	<b>K</b> L <b>E</b> D <b>G</b> D <b>R</b> F <b>W</b> W	<b>E</b> N <b>E</b> G <b>V</b> F <b>S</b> M <b>Q</b> Q	<b>R</b> Q <b>A</b> L <b>A</b> Q <b>I</b> S <b>L</b> P	<b>R</b> I <b>I</b> C <b>D</b> N <b>T</b> G <b>I</b> T	<b>T</b> V <b>S</b> K <b>N</b> N <b>I</b> F <b>M</b> S	720
721	<b>N</b> S <b>Y</b> P <b>R</b> D <b>V</b> N <b>C</b>	<b>S</b> T <b>L</b> P <b>A</b> L <b>N</b> L <b>A</b> S	<b>W</b> R <b>E</b> A <b>S</b>						745

The matched peptides cover 16% (125/745 AA's) of the protein.

Protein Name: Mutant MPO

Species: HUMAN

PN.user Accession #: 2

MS Digest Index #: 2

Masses are:

pI of Protein: 9.19

Protein MW: 83842.1 Da

Amino Acid Composition: A57 C17 D36 E35 F31 G45 H7 I30 K22 L87 M23 N41 P55 Q33 R66 S47 T39 V43 W12 Y19

MS Digest - Agilent Spectrum Mill Rev. 3.3.078

Supporting material 7 Van Antwerpen et al.: Peptide mapping of the four different MPO after enrichment on ConA column. Mapping was done by LC-ESI-MSMS and the protein summary was generated with Spectrum Mill ® (Agilent, Palo Alto, CA USA); **A** untreated recombinant myeloperoxidase; **B** recombinant myeloperoxidase incubated for 5 days at 37 °C in the absence of deglycosylation enzymes. **C** recombinant myeloperoxidase incubated for 5 days at 37 °C in the presence of deglycosylation enzymes. Glycosylation sites are in green and recorded peptides in red. The MPO sequence starts with the propeptide sequence. **D** Recombinant mutant myeloperoxidase (Q257→T) devoid of heme retaining the six N-glycosylation sites.