

Diverse proteins homologous to inositol monophosphatase

Andrew F. Neuwald¹, John D. York² and Philip W. Majerus²

¹*Institute for Biomedical Computing, Washington University School of Medicine, 700 South Euclid, Box 8036, St. Louis, MO 63110, USA* and ²*Division of Hematology-Oncology, Washington University School of Medicine, 660 South Euclid, Box 8125, St. Louis, MO 63110, USA*

Received 27 August 1991; revised version received 24 September 1991

Bovine inositol monophosphatase (IMP) and several homologous proteins were found to share two sequence motifs with bovine inositol polyphosphate 1-phosphatase (IPP). These motifs may correspond to binding sites within IMP and IPP for inositol phosphates or for lithium, since both substances are bound by these proteins. This suggests that the proteins homologous to IMP, which have diverse biological roles but whose function is not clear, may act by enhancing the synthesis or degradation of phosphorylated compounds.

Amino acid sequence; Inositol phosphate; Signaling pathway

We have found several proteins homologous to the inferred amino acid sequence of bovine inositol monophosphatase (IMP) [1], an enzyme of the inositol phosphate second messenger signaling pathway [2].

A search of the PIR protein database (release 28.0) using the FASTA program of Pearson and Lipman [3] yielded homologies between IMP and 2 inferred *Escherichia coli* proteins: the products of the *suhB* and *amtA* genes, mutations which result in enhanced synthesis of the heat shock transcription sigma factor [4], and deficiency in ammonium transport [5], respectively. Homologies between IMP and the products of the *Neurospora crassa qa-x* [6] and *Aspergillus nidulans qutG* [7] genes, which are both part of a cluster of genes involved in quinic acid utilization, have been reported previously [1]. A search of the GenBank database (release 67) using the FASTA program [3] uncovered a fifth homology to an open reading frame (ORF) upstream from the *pss* gene of *Rhizobium leguminosarum* [8] which is required for exopolysaccharide (EPS) synthesis and nodulation of peas. (Only the 3' end of the sequence is known since the 5' end of this ORF was deleted during cloning of the fragment.)

A well conserved motif within these proteins, defined as W- \bar{x} -[IV]-D-P-[IL-D-G-T- \bar{x} (2)]-[FY]- \bar{x} -[HK] (motif A, Fig. 1), was compared to sequences in the PIR and the translated GenBank databases using a UNIX lexical analysis program [9]. At most, 1 insertion or deletion at a variable residue (\bar{x}) and 1 substitution at a consensus

residue was allowed. An exhaustive search yielded only 1 additional protein with this motif, bovine inositol polyphosphate 1-phosphatase (IPP) [10], which also contains a second motif defined as W-D- \bar{x} (2)-[AG]-[AG]- \bar{x} -[AIL]-[ILV]-[ALV]- \bar{x} (3)-G-[AG] (motif B). Based on differences in the consensus pattern when the IPP sequence is included, motif A was redefined as W-x(0,1)-[IV]-D-P-[IL]-D-x-T-x(2)-[FY]-x-[HK]. These motifs may correspond to binding sites within IPP and IMP for inositol phosphates or for lithium since both proteins are inositol phosphate phosphatases and are uncompetitively inhibited by lithium (see references in [2,10]).

What do these homologies tell us? The extensive homology to IMP and the presence of 2 motifs similar to patterns found in IPP suggest that the homologous proteins are related to these phosphatases. Perhaps these proteins are components of a signaling pathway. The findings that *suhB* is probably identical to the *ssyA* gene [4], which was identified as an extragenic suppressor of a mutation affecting protein secretion, and that *amtA* is identical to the *cysQ* gene, which is required for efficient biosynthesis of cysteine (Neuwald et al., unpublished), suggest a pleiotropic role for these genes. This, along with preliminary evidence linking the *qa-x* gene to catabolite repression [11], raises the possibility that these proteins act by enhancing the synthesis or degradation of phosphorylated messenger molecules as adenylate cyclase and phosphodiesterases act in the synthesis or degradation of cyclic AMP, a signal for catabolite repression in *E. coli*.

Correspondence address: A.F. Neuwald, Institute for Biomedical Computing, Washington University School of Medicine, 700 South Euclid, Box 8036, St. Louis, MO 63110, USA. Fax: (1) (314) 3625872.

Acknowledgement: A.F.N. is a trainee in Medical Informatics – National Library of Medicine Training Program Grant 5-T-LM07049.

MADP ----- WQECMDYAVTLAGQAGEVVREALK ----- NEM	(1-31)	IMP
MTSRTTTTATELDEIYTF AVQLGKDAGNLLMEARLRFSNNNANHDKESTTQ	(1-51)	Qa-X
MDCP-IPQTELDEIYAFATDLARKAGQLLLERVN ----- DRNS-EQ	(1-39)	QutG
M ----- HPMLNI AVRAARKAGNLI AKNYE ----- TPD	(1-27)	SuhB
----- MLDQVCQLARNAGDAIMQVYD ----- GTK	(1-24)	AmtA
----- motif A -----		
SILTD ----- NPTWIIDPIDGTTNFVHGFPPVAVSIGFVVNKKMEFGIVYS	(79-124)	IMP
PYLVTHTTPTWVVDPLDGTVNYTHLFP MFCSIAFLVDGTPVIGVICA	(102-149)	Qa-X
EYLIDE-QPTWC VDPLDGTVNFTHAF PMFCSVIGFIVNHYPVIGVIYA	(88-134)	QutG
EGTDQ ----- DVQWVIDPLDGTTFIKRLPHFAVSI AVRIKGRTEVAVVYD	(73-118)	SuhB
EVRRQHW-QRYWLVDP LDGTKEFIKRN GEFTVNIALIDHGKPI LGVVYA	(71-117)	AmtA
W-VDPIDSTYQYIK	(151-163)	IPP
----- motif B -----		
CLEDKMYTGRKGKGAFCNG-QKLQVSHQ ----- EDITKSLLVTELGSS	(125-166)	IMP
PMLGQLFTACKGRGAWLN ETQRLPLVRQ ----- PMPKSAPGGCVFSC EWGKD	(150-196)	Qa-X
PMLNQLFSSCINRGAWLNEMQQLPLIRKPSIPPLPATAPSKCIFACEWGKD	(135-185)	QutG
PMRNE LFTATRGQGAQLNGYRL LGSTAR ----- DL DGTILATGFP-FK	(119-160)	SuhB
PVMNVMYSAAE GKAWKE ECGVRKQIQVR ----- DARPP LVVIS ----- RSH	(118-158)	AmtA
RTPETVRIILSNIERLLCLP ----- IHGIRGVGTAALNMCLVAA	(167-205)	IMP
RKDRPEGNLYRKVESFVNMAAEV GGRGKGGMVHGVRSLGSATLDLAYTAM	(197-247)	Qa-X
RRDIPDGT LQRKIESFVNMAAERGSRGKGGMVHGVRSLGSATMDLAYTAM	(186-236)	QutG
AKQYATTYINIVGKLFNEC ----- ADFRRTGSAALDLAYVAA	(161-197)	SuhB
ADAE ELKEYLQQLGE ----- HQTT SIGSS-LKFCLVAE	(159-189)	AmtA
SFLADHAI ----- PKCTNIGSS-LKFCLLAE	(ORF)	
----- motif B -----		
GAADAYYEMG-IHCWDV VAGAGIIVTEAGGVLLDV ----- TGGPFD	(206-244)	IMP
GSFDIWWEGG-CWEWDVAAGIAILOEAGGLITSANPPEDWATAEIPD	(248-293)	Qa-X
GSVDIWWEGG-CWEWDVAAGIAILLEAGGLVTAANPPED-IEGPIEP	(237-281)	QutG
GRVDGFFEIG-LRPWDF AAGELLVREAGGIVSDF ----- TGGH-N	(198-235)	SuhB
GQAHVYPRF GPTNIWDTAAGH AVAAAAGA HVHDW ----- QKPLD	(190-229)	AmtA
GKADVYPRFTRTMEWDTAAGD AVLRAAGGSTVTL ----- DGTPLT	(ORF)	
WDS CAAHAILRAMGG	(316-330)	IPP
----- motif B -----		
----- LMSRRVIAS-SNKT LAERIAKEI-QIIP LQRDDED -----	(245-277)	IMP
VKLGSRLYLVRPAGP SEGETAREGQERTIREVWRRVRALDYTRPGA ---	(294-340)	Qa-X
VKLGSRLYLAI RAGPSETETGRETQERTVREWRRVRQLDYERPTROS	(282-330)	QutG
----- YMLTGNIVA-GNP ----- RVVKAM-LANMRDELS DALKR	(236-267)	SuhB
----- YTPRESFLN-PGF ----- RVSTY -----	(230-246)	AmtA
----- YGKTGTAD ----- FDFANPNFISWGG RKRVLEPA -----	(ORF)	

Fig. 1. Conserved regions between inositol monophosphatase and homologous proteins. Similar residues are bold. Comparison of bovine inositol monophosphatase (IMP) to the products of the *Neurospora crassa qa-x* gene (Qa-X) [6], the *Aspergillus nidulans qutG* gene (QutG) [7] and the *Escherichia coli suhB* (SuhB) [4] and *amtA* [5] genes (AmtA), and to the C-terminal peptide fragment inferred from the 3' end of an ORF upstream from the *Rhizobium leguminosarum pss* gene [8] yielded homology scores of 27, 35, 43, 21 and 10 s.d. units, respectively. Additional comparisons yielded homology scores of 139 (QutG, Qa-X), 18 (SuhB, Qa-X), 23 (SuhB, QutG), 13 (AmtA, Qa-X), 16 (AmtA, QutG) and 28 (AmtA, SuhB) s.d. units. Scores were calculated by computing optimized alignments of one of the protein sequences with 1000 random permutations of the other sequence using the RDF2 program of Pearson and Lipman [3]. IMP and bovine inositol polyphosphate 1-phosphatase (IPP) are not homologous (score: 2.5 s.d. units) but they share 2 motifs (A and B).

REFERENCES

- [1] Diehl, R.E., Whiting, P., Potter, J., Gee, N., Ragan, C.I., Linemeyer, D., Schoepfer, R., Bennett, C. and Dixon, R.A.F. (1990) *J. Biol. Chem.* 265, 5946-5949.
- [2] Majerus, P.W., Connolly, T.M., Bansal, V.S., Inhorn, R.C., Ross, T.S. and Lips, D.L. (1988) *J. Biol. Chem.* 263, 3051-3054.
- [3] Pearson, W.R. and Lipman, D.J. (1988) *Proc. Natl. Acad. Sci. USA* 85, 2444-2448.
- [4] Yano, R., Nagai, H., Shiba, K. and Yura, T. (1990) *J. Bacteriol.* 172, 2124-2130.
- [5] Fabiny, J.M., Jayakumar, A., Chinault, A.C. and Barnes Jr., E.M. (1991) *J. Gen. Microbiol.* 137, 983-989.
- [6] Geever, R.F., Huiet, L., Baum, J.A., Tyler, B.M., Patel, V.B., Rutledge, B.J., Case, M.E. and Giles, N.H. (1989) *J. Mol. Biol.* 207, 15-34.
- [7] Hawkins, A.R., Lamb, H.K., Smith, M., Keyte, J.W. and Roberts, C.F. (1988) *Mol. Gen. Genet.* 214, 224-231.

- [8] Borthakur, D., Barker, R.F., Latchford, J.W., Rossen, L. and Johnston, A.W.B. (1988) *Mol. Gen. Genet.* 213, 155–162.
- [9] Lesk, M.E. and Schmidt, E. (1986) in: *UNIX Programmers Manual* (Computer Systems Research Group, University of California, Berkeley) Supplementary Documents vol. 1:16, USENIX Association, Berkeley, California.
- [10] York, J.D. and Majerus, P.W. (1990) *Proc. Natl. Acad. Sci. USA* 87, 9548–9552.
- [11] Giles, N.H., Geever, R.F., Asch, D.K., Avalos, J. and Case, M.E. (1991) *J. Hered.* 82, 1–7.