Biology of Inositols and Phosphoinositides

Subcellular Biochemistry
Volume 39

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Subcellular Biochemistry Volume 39

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Preface

From being to becoming important, *myo*-inositol and its derivatives including phosphoinositides and phosphoinositols involved in diversified functions in wide varieties of cells overcoming its insignificant role had to wait more than a century. *Myo*-inositol, infact, is the oldest known inositol and it was isolated from muscle as early as 1850 and phytin (Inositol hexakis phosphate) from plants by Pfeffer in 1872. Since then, interest in inositols and their derivatives varied as the methodology of isolation and purification of the stereoisomers of inositol and their derivatives advanced. Phosphoinositides were first isolated from brain in 1949 by Folch and their structure was established in 1961 by Ballou and his coworkers. After the compilation of scattered publications on cyclitols by Posternak (1965), proceedings of the conference on cyclitols and phosphoinositides under the supervision of Hoffmann-Ostenhof, were published in 1969. Similar proceedings of the second conference on the same subject edited by Wells and Eisenberg Jr was published in 1978. In that meeting at the concluding session Hawthorne remarked "persued deeply enough perhaps even myoinositol could be mirror to the whole universe". This is now infact the scenario on the research on inositol and their phosphoderivatives. Finally a comprehensive information covering the aspects of chemistry, biochemistry and physiology of inositols and their phosphoderivatives in a book entitled Inositol Phosphates written by Cosgrove (1980) was available. Inositol Metabolism in Plants edited by Morre, Boss & Loewus, was published in 1990. In 1996 a special volume of Subcellular Biochemistry (Vol 26) entitled "*Myo*-inositol Phosphates Phosphoinositides and Signal Transduction' edited by Biswas and Biswas was brought out to record the explosion of interest due to discovery of the "phosphoinositide effect". It is thought pertinent to publish another volume of Subcellular Biochemistry taking into an account of the advancement of knowledge in this area during last decade or so and this volume on "Biology of Inositol & Phosphoinositides" is the outcome of the present theme.

Implicitly what is to be mentioned that this volume is not intended to be all inclusive. However, this is aimed at giving a wide coverage starting from the structural aspect of Inositols and their derivatives to functional genomics, genetics of Inositol metabolism and storage; phosphoinositide metabolism in health and disease, in stress signaling and finally evolutionary consideration on the basis of genomics of diversified organisms. Repetitions as appear in some chapters have been retained because of the interest of the similar problems tackled from different angles using different systems following specific pattern of presentation and elucidation of importance of inositols and its derivatives in the diversified cellular functions.

Inositols, in particular *myo*-inositol, plays a central role in cellular metabolism. An array of complicated molecules that incorporate the inositol moiety are found in nature. Structural heterogeneity of inositol derivatives is compounded by the presence of stereo-and regio-isomers of the inositol moiety. Because of the large number of isomeric inositols and their derivatives present in nature, a detailed understanding of the structural, stereochemical and nomenclatures related issues involving inositol and its derivatives is essential to investigate biological aspects. A pertinent discussion of the stereochemical, conformational, prochiral, chiral and nomenclature issues associated with inositols and structural variety of inositol derivatives is presented. Murthy (Chapter 1) has taken painstaking effort in removing the confusion still present in the literature regarding the structure and nomenclature of inositol phosphates, phosphoinositides and glycosyl phosphoinositols. As far as possible along with old nomenclature the new assignment as recommended is also included to remove the confusion about the structural configuaration of inositol phosphates and phosphoinositides.

Loewus (Chapter 2) reviewed extensively *myo*-inositol biosynthesis from Glucose-6-phosphate and its catabolism particularly in plants related to cell wall biogenesis based on experimental data thus far available. Interestingly, *myo*-inositol is synthesized by both eukaryotes and prokaryotes specifically by two enzyme systems, one is M1P synthase and the other is IMP-phosphatase. Once MI is formed it is utilized for many biosynthetic processes in plants including formation of raffinose series of oligosaccharides, biosynthesis of isomeric inositols & their O-methyl ethers and membrane biogenesis. In addition to processes mentioned wherein the inositol structure is conserved, a major catabolic process competes for free MI and it is oxidized to D-Glucuronic acid by the enzyme MIoxygenase (MIOX). D-Glucuronic acid is subsequently converted to D-Glucuronic acid-1-P and UDP-D-Glucuronic acid by GlcUA-1-kinase and GlcUA-1-P uridyl –transferase respectively. UDP-D-GlcUA is the starting point to produce uronosyl and pentosyl component of cell wall polysaccharides. UDP-D-GlcUA was also generated directly from Glc-6-P through Phosphoglucomutase, UTP-Glc-1-P uridyl transferase and UDP-Glc dehydrogenase. Thus an alternative pathway to these cell wall polysaccharides that bypassed UDP-Glc dehydrogenase is recorded. These two pathways are now

referred to as the MIOP and SNOP. MI as potential precursor of L-tartarate and oxalate which are linked with L-ascorbate breakdown products has been recently suggested. It is expected that efforts will continue to uncover new molecular and biochemical details of the MIOP and SNOP interrelationship as well as linkage of ascorbic acid biosynthesis and MI metabolism in plants.

The functional genomics of *myo*-inositol metabolism is the new aspect where Tora-Sinajad and Gillaspy (Chapter 3) have detailed the MIPS, IMP and MIOX genes and proteins with an overall focus on determining gene function involved in the inositol anabolic and catabolic pathways in different systems. One central theme what emerges at present is that the genes encoding enzymes of these pathways are present in prokaryotes, unicellular and multicellular eukaryotes. However, the regulations and contribution to specific end products are different. Not only prokaryotes utilize *myo*-inositol in different pathways from yeast, plants or animals but also their genetic diversity for these genes differs. More interestingly, plants appear to exhibit more complexity with respect to the numbers of genes that encode MIPS, IMPase and MIOXase enzymes as well as other regulatory proteins. In prokaryotes, the focus has been onto cell wall and RNA processing.

Genetical studies of *myo*-inositol phosphates and phosphoinositides are at present at an initial stage and the progress recorded has been slow due to difficulties in raising the mutants with respect to metabolism of *myo*-inositol and its phosphoderivatives. A thought-provoking discussion on this aspect has been initiated by Raboy and Bowen (Chapter 4). They started in sequence *myo*-inositol and *myo*-inositol-hexakis-phosphate as focal points for the purpose of metabolism and functions along with evolutionary consideration on the basis of genetics and genomics data available for diversified organisms. Besides participation in signal transduction, the involvement of *myo*-inositol phosphates and phosphoinositides in other functions of basal cellular metabolism and housekeeping, is considered. Differences between divergent species with respect to *myo*-inositol phosphate and phosphoinositide pathways when analyzed through mutations that block specific sites often take alternative metabolic routes to provide that component leading probably to metabolic balancing. Finally, an understanding of how *myo*-inositol phosphates and phosphoinositides are compartmentalized has been elucidated.

Roberts (Chapter 5) has contributed ably about Inositols in Bacteria & Archaea giving in details the identity of varied *myo*-inositol compounds their enzymology and functions including infectivity and virulence. In fact, *myo*-inositol compounds are not ubiquitous in bacteria but restricted to certain classes of these organisms and surprisingly not involved in signal transduction pathways in any of those organisms thus far studied signifying that *myo*-inositol is required for other functions.

A considerable progress has been made in the recent past in genetic regulation of MIP synthase in yeast initiated by isolation of the first *S. cerevisiae* inositol auxotrophs and the subsequent cloning and sequencing of its structural gene INO1. Indepth studies on the regulation of INO1 revealing the complex

mechanisms controlling phospholipid metabolism related to cellular signaling pathways have been described succinctly by Nunez and Henry (Chapter 6).

Geiger (Chapter 7) dealt on the structure & mechanism of MIPS and compared the sequence alignment of MIPS from *S.cerevisiae*, *Mycobacterium tuberculosis* and *Archaeoglobus fulgidus*. When the yeast enzyme is aligned with the other two, there are significant differences in domain architecture. Enzymes from other two sources do not contain N-terminal 65 amino acids. However, both of them have C-terminal regions similar to yMIPS that serve to fix the relative orientation between the catalytic domain and Rossmann fold domain. The active site of MIPS is located between the bottom of the Rossmann fold domain and the beta sheet of the catalytic domain. A reasonable hypothesis for the detailed mechanism of the reaction has been discussed taking into consideration of the combination of the inhibitor-bound structure and the modeling approaches. Many significant questions raised remain to fully characterize the mechanism of MIPS in future. The combination of structural, biochemical and genetical studies on enzymes from different sources is leading to the complex mechanism for the conversion of G-6-P to MIP catalyzed by MIP synthase.

The review on phosphoinositide metabolism to understand the subcellular signaling in an organism and the functional coding of phosphoinositide signals deviation specifically in plants is attractive. Inositol phospholipids have multiple effects on cellular metabolism regulating cytoskeletal structure, membrane associated enzymes, ion channels and pumps, vescicle trafficking as well as producing second messenger. Boss, Davis, Im, Galvvo and Perera (Chapter8) discussed several aspects such as lipid-protein interactions, association of subcellular structure with inositol lipids domains, cellular pools of phosphatidylinositol 4,5 bisphosphate in order to understand subcellular signaling network in stress conditions.

In addition, Zonia and Munnik (Chapter 9) dealt with the functional coding of phosphoinositide signals during plant stress taking into consideration the pertinent discoveries on phosphoinositide signaling during cellular homeostasis, difference in phosphoinositide synthesis in different systems or their direct and indirect involvement in eliciting signals in unstressed cells and during both biotic and abiotic stress conditions including plant cell swelling and shrinking process. It is also apparent from discussions therein that the plant signals and cellular responses may differ in variety of ways from those in other organisms. Therefore, more and more new data about phosphoinositide signaling in plants are emerging which has been very ably presented.

A wider perspective on inositols and their metabolites in abiotic and biotic stress responses has been documented by Taji, Takahashi and Shinozaki (Chapter 10). Inositol and its metabolites function as both osmolytes and secondary messengers under biotic and abiotic stresses. The accumulation of different osmolytes during osmotic stress is an ubiquitous biochemical mechanism found in different organisms from bacteria, fungi and algae to plants and animals. Plants accumulate many types of inositol derivatives during abiotic

stresses such as drought, low temperature and salinity in contrast most animals accumulate only *myo*-inositol. They have dwelt on molecular basis of osmolyte strategies in animals and plants, *myo*-inositol 1-phosphate, D-ononitol and D-pinitol, galactinol and reffinose as osmolytes in plants, inositol phosphates as signaling molecules with special reference to inositol (1,4,5) trisphosphate levels in response to abiotic stress, involvement of inositol trisphosphate in abscisic acid signaling, enzymes that regulate inositol trisphosphate levels and finally inositol hexakisphosphate as a signaling mediator with special reference to mRNA export, DNA repair, DNA recombination, vasicular trafficking, antioxidants and antitumor compounds. Functional aspects of inositols and its phosphoderivatives thus far mentioned once again support the view that they play a central role in cellular metabolism.

The important role of Inositol phosphate and phosphoinosities in health & disease has been critically tackled by Shi, Azab, Thompson and Greenberg (Chapter 11). They discussed the involvement of Ins(1,4,5)P_3 in neurological disorders such as Bipolar and Alzheimer's diseases. A correlation of abnormal function of IP_3R1 has been found associated with epilepsy and ataxia in mice as well as Huntington's disease in human patients. Moreover alteration in InsP3/Ca^{2+} signaling is one of the suggested mechanisms for malignant hyperthermia in humans. The Ca^{2+} overload due to the increased $InsP_3$ activity was suggested as a major contributor to the severe cardiac arrhythmias seen during the ischemia/reperfusion cycles. $InsP_6$ has been shown to exhibit strong antioxidant properties and is used as a potential anti-neoplastic therapy. In addition, they dwelt on the diseases caused by perturbation of PI metabolism such a P13K/AKT pathway in cancer and in type 2 diabetes as well as P15P in insulin signaling. P1(4,5) P_2 accumulation due to deficiency of OCRL1 gene (Oculo-Cerebro-Rinal syndrome of Lowe) is consistent with loss of function of OCRL1 product [P1(4,5)P2–5 phosphatase] along with other diseases associated with the myotubularin family opening up possibilities for effective drug design.

Mammalian *myo*-inositol 3-phosphate synthase (MIPS) and its role in biosynthesis of brain inositol and its clinical use as psycho-actve agent has been documented by Parthasarathy, Seelan, Tobias, Casanova and Parthasarathy (Chapter 12). They tried to draw attention to inositol homeostasis in mammalian brain by inositol synthase through dietary intake of inositol and continuous hydrolysis of inositol monophosphate by IMPase1. Inositol pathways have been implicated in the pathogenesis of bipolar disorder, with the mood stabilizers, valproate and lithium targeting inositol synthase and IMPase1 respectively. The inhibitory effect of valproate on inositol synthase suggests that this enzyme may be a potential therapeutic target for modulating brain inositol levels. Clinical studies on panic disorder, schizophrenia, obsessive –compulsive disorder, post traumatic stress disorder, attention deficit disorder, autism and alzheimer's disease by supplying inositol to the patient, have been presented implicating the biochemical and genetical regulation of inositol synthase and IMPase in the brain.

Presence of MIPS throughout evolutionary diverse organisms from eubacteria to higher eukaryotes presupposes its early origin. Analysis by multiple sequence alignment, phylogenetic tree generation and comparison of crystal structures thus far available provide new perspective into the origin and evolutionary relationships among the various MIPS proteins. Evolutionary divergence of MIPS and significance of a "core catalytic structure" have been succinctly discussed by GhoshDastidar, Chatterjee, Chatterjee and Lahiri Majumder (Chapter 13). Finally the interesting question arises whether acetolactate synthase homologue in *E.coli* can function as a MIPS present in *Synechocystis* which lacks in similar structure to the known MIPS from other prokaryotic and eukaryotic sources so far compared.

It is thus apparent from foregoing discussion that the importance has been given on the metabolism of inositol and phosphoinositides in different organisms from archaea to mammals aiming at working out metabolomics and to unravel a variety of network of metabolism. Attention has also been focussed on the key enzyme MIPS (*myo*-inositol–3-phosphate synthase), its structure, sequence, mechanism of action and functional genomics, purifying it from variety of organisms such as *Archaeoglobus fulgidus, Mycobacterium tuberculosis, Saccharomyces cerevisiae, Porteresia coarctata* and mammals. The alignment of sequences of this enzyme from different sources helps in working out diversification and evolution of *myo*-inositol–3 phosphate synthase and proposition of different models for its action mechanism. The important role of *myo*-inositol, its phospho-derivatives and phospholipids in health and disease is an emerging aspect which warrant significant attention at this time to work out their implications in pharmacogenomics. The role of *myo*-inositol and its phospho-derivatives under stress conditions opens up a new vista in functional genomics to identify the variety of gene expression under a particular condition in different systems and genetic along with molecular genetic studies have provided a thought provoking discussion implicating inositol phosphates and phosphoinositides functions in signaling other than that in signal transduction in mammals. Finally, the framework of molecular evolution of MIPS from archea to man has been proposed taking into consideration of sequence alignment, comparison of crystal structures and the sequence homology of the core catalytic domains of MIPS. Similar studies on the sequence deviations in IMPase and phytase from variety of organisms may be initiated in future. Our endeavour in organizing this volume of Subcellular Biochemistry will be fruitful if the workers in this emerging field embracing different disciplines such as molecular biology, chemistry, bioinformatics, psychiatry, agriculture, medicine, microbiology and biotechnology are benefitted from the information compiled by the experts in the field of biology of inositols and phosphoinositides.

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