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EDITORIAL

Polyamines and transglutaminases: future perspectives

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Preface

This is the fifth special issue focused on Polyamines available on Amino Acids. It is an editorial initiative on the occasion of the international meeting held in Wien (Austria) in 2015. Some articles in this issue highlight work on Transglutaminases (TGases). The first edition, "Polyamines and their analogs in cancer and other diseases", was previously published in this journal (Editorial, 2007, Amino Acids 33:173) which covered topics discussed at the 1st international polyamine meeting held in Tivoli (Rome) in 2006. The 2007 issue was co-edited with a prominent polyamine field expert, Professor Kazuei Igarashi, as guest editor and dedicated to the memory of Professor Nikolaus Seiler, an authoritative scientist who had enormously contributed to the polyamine research. The second special issue entitled, "Polyamines in Biological Systems" published in Amino Acids (Vol 38, no. 2, 2010), covered the 11th International Congress on Amino Acids, Peptides and Proteins held in Wien in 2009, organized by Professor Gert Lubec and myself. The 'editorial' tradition on the occasion of the 2nd International Conference of Polyamines in cancer and other diseases, held at Tivoli (Rome) in 2010, was continued with a third special issue on the "Role of polyamines, their analogs and transglutaminases in biological and clinical perspectives", edited by Professor Igarashi and myself (Amino Acids, Vol 42, numbers 2–3, 2012). Research developments in the areas of biosynthesis and action of the ubiquitous polyamines, with particular focus on the biology of normal and cancerous eukaryotic cells, also covered the field in plants, represented the theme of the 4th special issue on Polyamines and Transglutaminases: Biological, Clinical and Biotechnological Perspectives (published in Amino Acids, Vol. 46, no. 3, March 2014). These previous successes became the basis and the driving force for holding the 2012 International Congress on "Polyamines Biological and Clinical Perspectives" in the marvelous city of Istanbul, Turkey at Istanbul Kultur University, organized by Professor Narcin Palavan Unsal and myself.

This Special Issue of Amino Acids brings together 17 peer-reviewed manuscripts covering the essence of the lectures and posters, including subjects of discussion, presented at the International Congress on Amino Acids, Peptides and Proteins, held in Wien, August 3–8, 2015, and organized by Professor Gert Lubec, Editor in Chief-of-Amino Acids, with myself covering the "Polyamines Session". In addition, it covered additional articles on the physiological roles of polyamines, which were solicited from other experts who could not attend the meeting. The articles deal with high-class research covering progress up to the end of 2015, and contain literature references, figures, tables and reaction schemes. In fact, the issue is intended to provide a relatively short overview of some important concepts and notions mainly on biogenic amines, including polyamines, and represents an important tool available to new and old players in this intriguing field.

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All manuscripts in this special issue were subjected to external peer reviewing according to the policy of this journal.

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As mentioned above, posttranslational modifications of proteins via covalent binding of polyamines with special emphasis on transglutaminases (TGases) and hypusine biosynthesis are also highlighted in this issue. It is worthwhile mentioning here that an excellent edited book on "Transglutaminases" (by Hitomi, Kojima and Fesus) was published in 2015 by Springer Japan, which featured a number of review articles on this polyhedral family of enzymes.

Editorial

This issue is focused on biogenic amines, the polyamines, an editorial initiative covering research findings on the biological roles of polyamines and transglutaminases (TGases), most of which were deliberated at the 2015 international meeting held in Wien, Austria. Polyamines, i.e., spermine, spermidine and putrescine, are ubiquitous molecules essential for the physiological processes, including cell growth, development and differentiation. The diamine putrescine is biosynthesized from ornithine by ornithine decarboxylase (ODC) and from arginine via a couple of intermediates initial enzyme arginine decarboxylase (ADC). Through successive reactions, putrescine is converted to the triamine spermidine by spermidine synthase and tetramine spermine by spermine synthase, involving the transfer of an aminopropyl moiety from decarboxylated S-adenosylmethionine. Polyamines exist as protonated polycations at physiological pH in living cells and interact with nucleic acid, acid proteins and phospholipids. Numerous studies emphasize important activities exhibited by polyamines, including activation of kinases involved in signal transduction, regulation of ion channel gating, and modulation of oxidative processes (Neel and Tonks 1997; Ficker et al. 1994). Moreover, several investigations have highlighted a number of biochemical processes in which polyamines exert a regulatory function (Gerner and Meyskens 2004; Pegg 2009; Igarashi and Kashiwagi 2010). Thus, polyamines play mandatory roles in several cell functions, such as DNA synthesis, proliferation, gene transcription regulation, translation, posttranslational modification, membrane stability/function, apoptosis, cytoskeleton function, modulation of cell cycle, and ion channeling (Pegg and Casero 2011; Ramani et al. 2014). The role of polyamines in the RNA world include translational regulation (Igarashi and Kashiwagi 2010) and activation of the eukaryotic initiation factor 5A (eIF5A) by spermidine (Lee et al. 2011). A large number of physiological functions at the levels of biosynthesis, metabolism and transport are attributed to polyamines. Therefore, the field is complex and understanding the physio-pathological consequences associated with the variations in their cellular concentration is a challenging task.

In eukaryotes, one of the important functions of polyamine spermidine is being the precursor of hypusine [Nε-(4-amino-2-hydroxybutyl)lysine], an unusual basic amino acid that also post-translationally modifies the eukaryotic initiation factor eIF5A. This modification is essential for eIF5A activity and protein synthesis during the elongation step (Childs et al. 2003; Park 2006; Hyvönen et al. 2007; Park et al. 2010). The amino acid hypusine derives from the addition of the 4-aminobutyl moieties to lysine; this reaction is catalyzed by deoxyhypusine synthase, which uses spermidine as substrate (Park 2006).

Cellular polyamine concentrations are highly regulated. Intracellular accumulation of polyamines, due to either high extracellular levels or deregulation of the systems that control polyamine homeostasis, can induce programmed cell death (or apoptosis) in various cell types (Tobias and Kahana 1995; Pignatti et al. 2004; Seiler and Raul 2005). Therefore, maintenance of the proper concentration of polyamines is necessary for above functions, whereas excess in their levels could lead to toxicity (Toninello et al. 2004; Amendola et al. 2009; Pegg 2013). Toxic effects are also a result of polyamine catabolism due to generation of reactive oxygen species (ROS) by a variety of oxidases. Some of these, such as diamine oxidase and serum amine oxidase, are well known, whereas others, such as polyamine oxidases and spermine oxidase (SMO), have only recently been isolated. The mechanistic aspects of these oxidase activities, excluding SMO, are known, and their cofactors and preferred polyamines as substrates have been characterized. Amine oxidases can be classified as Cu^{2+} containing (or semicarbazide-sensitive), such as diamine oxidases, vascular-adhesion protein-1 and lysiloxidases, and FAD-dependent AOs (including monoamine, polyamine, and spermine oxidases; see Table 1 in Agostinelli et al. (2010).

As often reported, spermine and other polyamines are transported in mammalian mitochondria by an energy-dependent mechanism, with electrical membrane potential $\Delta\Psi$ as the driving force (Toninello et al. 2004). As a result of this transport, spermine exhibits several significant effects for mitochondrial bioenergetics functions (Grancara et al. 2014), particularly at the level of the mitochondrial permeability transition (MPT), a phenomenon related to intrinsic apoptosis. Spermine uptake into the mitochondrial matrix also indicates the need to identify a catabolic pathway in these organelles. Polyamine oxidase activity is present in rat tissues, particularly in liver, in which the highest activity was detected in the light mitochondrial fraction (Pavlov et al. 1991), most of the lysosomes and peroxisomes and intact Golgi membranes. However, the first preliminary kinetic characterization of a copper-containing (semicarbazide-sensitive) amine oxidase in the liver mitochondrial matrix was reported in 2009, with putrescine and spermine as substrates (Cardillo et al. 2009). Among

the polyamine oxidases belonging to the family of flavoprotein amine oxidases, spermine oxidase (SMO) is highly specific to spermine as substrate producing spermidine, H_2O_2, and 3-aminopropanal with no prior acetylation step (Wang et al. 2001, 2003; Cervelli et al. 2012). Using purified bovine liver mitochondria, the capacity for natural polyamine uptake was verified. A kinetic approach was used to determine the presence of an MDL 72527-sensitive enzyme with spermine oxidase activity in the matrix of bovine liver mitochondria. Immunoblot analysis of mitochondrial fractions and immunogold electron microscopy observations of purified mitochondria unequivocally confirmed the presence of a protein immunodecorated by anti-spermine oxidase antibodies in the mitochondrial matrix. These findings indicated that the bovine liver mitochondrial matrix contains an enzyme belonging to the spermine oxidase class (Bonaiuto et al. 2015). The observation that polyamines are transported in mitochondria by an energy-dependent mechanism (Toninello et al. 2004) thus reinforce the hypothesis that mitochondria contain the spermine catabolism enzyme. In addition, SMO, which is normally present in mammalian cells at low levels, is highly inducible by a variety of stimuli, such as *Helicobacter pylori* infection (in gastric cancer), the pro-inflammatory TNF-alpha (in lung epithelial cells) (Babbar and Casero 2006), and polyamine analogs (Wang et al. 2001); Devereux et al. 2003), indicating SMO is associated with several pathological conditions.

Polyamines are also substrates for TGases. TGases are ubiquitous enzymes with multifaceted functions. The presence of an acyl-enzyme thioester intermediate between an active site cysteine and a polypeptide-bound glutamine and further the reaction of the thioester intermediate with a suitable nucleophile is the basis of the enzymatic mechanisms (Folk and Cole 1966). Research on TGases includes both basic and applied aspects, involving the fields of medicine (e.g., development of diseases such as cancer and Alzheimer), plant science (e.g., regulation of processes such as photosynthesis and fertilization), and food and material sciences (reviews: Scarnato et al. 2016; Aloisi et al. 2016).

The conformational state of TG2 has been the object of intense scrutiny since the seminal work by Khosla's team (Stanford University) (Pinkas et al. 2007), showing that TG2 assumes two very different conformations upon binding Ca^{2+} and purine nucleotides. Redox changes have emerged as further potent regulators of TG2, as the oxidizing nature of the extracellular environment inactivates the TG2 enzyme via cysteine oxidation (Jin et al. 2011; Yi and Khosla 2016). Using hydrogen/deuterium exchange monitored by mass spectrometry, Sollid and Iversen (University of Oslo) have demonstrated that TG2-reactive antibodies, auto-antibodies which are highly specific to celiac disease (Iversen 2015), cause conformational changes in TG2 (Iversen et al. 2014). These findings open the possibility that celiac antibodies alter TG2 properties and its association with the TG2 interactome. The theme of protein aggregation by TG2 transamidation is a classic in the TG2 literature, and several studies have suggested that TG2 contributes to the accumulation of protein aggregates by cross-linking in neurodegenerative disorders. It was suggested that TG2 is involved in the clearance of ubiquitinated protein aggregates by autophagy, a cellular process responsible for the degradation of protein aggregates through the autophagosome–lysosome system (D'Eletto et al. 2012). The same group reported the involvement of TG2 also in the turnover and degradation of fragmented and damaged mitochondria (mitophagy) (Rossin et al. 2015), and observed that the absence of TG2 led to an accumulation of dysfunctional mitochondria. As a consequence of the wide implication of TG2 and family members in disease, there is a growing interest for effective inhibitors specific for TG2. This is necessary in all areas where aberrant expression or function of TG2 has been consistently observed in cell and clinical samples (e.g., fibrosis, celiac disease, cancer).

The first part of the issue has articles on polyamine catabolism and its role in animals. This information could be evaluated for possibility of targeting this metabolic pathway for new antiproliferative therapies in animals. Little is known about the molecular mechanism(s) of these amines in animal physiology. Polyamines are often present at high concentrations in rapidly dividing tumor cells and growing tissues, and likely active in hyperproliferative diseases such as in various cancer cells (Bachrach 2004). Therefore, special attention has been paid on their involvement in carcinogenesis and in developing new approaches to cancer therapy and other diseases. Amine oxidases (AOs) regulate the levels of polyamines and generate cytotoxic metabolites (Arancia et al. 2004; Agostinelli et al. 2004). Interestingly, spermine oxidase (SMO), a FAD-containing enzyme, specifically oxidizes spermine (Spm) and SMO dysregulation alters polyamine homeostasis, participating in the aetiology of several pathological conditions, including cancer (Amendola et al. 2009; Agostinelli et al. 2010; Casero and Pegg 2010). Direct mechanistic links between inflammation, SMO activity, ROS production and carcinogenesis have been demonstrated (Goodwin et al. 2008). Main biochemical, cellular and physiological processes in which SMO is involved has been highlighted (Cervelli et al. 2012). In this issue, the stability, thermal and chemical denaturation of SMO was compared with bovine serum amine oxidase (BSAO), a copper amine oxidase. Both were found to oxidize SPM. Homodimeric and monomeric SMOs were found catalytically active. The SMO mutant deprived of all but two internal residues, i.e., Cys263 and Cys429, according to molecular modeling analysis, was constructed and characterized in its monomeric form. High-sensitivity, differential scanning calorimetry of SMO

was carried out to analyse its thermal stability and then compared with that of BSAO, to gain an insight into the unfolding process (Cervelli et al. 2016).

That polyamine levels are higher in breast tumors is highlighted in the article by Thomas et al. (2016b) as compared to lower levels in the surrounding tissues (Cervelli et al. 2014). Estradiol, the female hormone implicated in the origin and progression of breast cancer, stimulates ODC and AdoMetDC (Thomas and Thomas 2001). Consequently, antiestrogens suppress polyamine biosynthesis. Tamoxifen is one of the most widely used antiestrogen for breast cancer treatment (Thomas et al. 2004). Endoxifen, an active metabolite of tamoxifen, in addition to inhibiting ODC and AdoMetDC, increases the activity of two aminoxidases, APAO and SMO.

Increased ODC causes putrescine synthesis from ornithine during the growth of various cancer cells and high polyamine levels are often seen in rapidly dividing tumor cells and growing tissues in addition to increased uptake of polyamines (Marton and Pegg 1995). In this issue Nowotarski et al. (2016) show that ODC is post-transcriptionally regulated by the RNA binding protein TTP. TTP is a destabilizing RNA binding protein that typically binds to labile mRNA and relocates them to mRNA decay machinery. Knocking out TTP results in the increased ODC mRNA stability as well as its enzyme activity, suggesting that TTP destabilizes the ODC mRNA. Additionally, the authors show that the 3′UTR of ODC indeed contains a TTP-regulated element. Thus, complexity of ODC regulation was revealed by demonstration of another trans-acting factor that affects ODC stability and expression.

Observations on the interactions of biogenic amines with mitochondria have revealed intriguing mechanisms of amine oxidation and transport (Agostinelli et al. 2004, 2010; Battaglia et al. 2010). In a minireview, Grancara et al. (2016a) discuss interactions of mitochondria with the amines, as well as a new morphology of the mitochondrial inner membrane and that of the cristae (Scorrano 2013), raising possibilities for other interpretations on their functions. Also, another phenomenon regarding membrane permeability specific to the outer membrane with a different mechanism from that induced in the inner membrane is discussed (Green and Reed 1998). Also, polyamines in mitochondria are discussed in relation to reactive oxygen species (ROS) production and possibilities for new therapeutic strategy against cancer.

In relation to spermine cycling in mitochondria, Grancara et al. (2016b) demonstrate the intriguing molecular mechanism of spermine efflux from energized mitochondria mediated by the adenine nucleotide translocase. This discovery, when considered together with the uptake mechanism of spermine (Toninello et al. 1988, 1992), suggests that spermine can cycle across the mitochondrial membranes. The rate of this cycling regulates the spermine concentration in the matrix and, consequently, its prevention or induction of mitochondrial permeability transition (Agostinelli et al. 2010).

Understanding the role of highly conserved proteins across evolution such as those involved in protein synthesis, is essential not only to establish the basis of the mechanisms involved in the regulation of gene expression but also to help modulate such processes during disease conditions. Eukaryotic initiation factor 5A (eIF-5A) is one of these proteins and constitutes an interesting drug target. eIF-5A is a regulatory protein that upon posttranslational modification controls translation and subsequent protein biosynthesis. In a minireview, Migliaccio et al. (2016) consider protein phosphorylation as a link between eukaryotic translation elongation factor 1A (eEF1A) and signal transduction pathway. Thus, different kinases that recognize the Ser and Thr residues of eEF1A1 and eEF1A2 isoforms regulate their function in different cellular processes such as cell survival and apoptosis. In this context, polyamines seem to play a role in the regulation of the translation elongation process through Ser/Thr phosphorylation of translation elongation factors.

Nakanishi and Cleveland (2016) consider that elevated levels of EIF5A connote poor prognosis for several human tumor types. Additionally, hypusinated eIF5A appears to be required for the growth and tumorigenic potential of several cancer cell lines. Notably, eIF5A seems essential for translation of proteins harboring polyproline motifs. Development of agents that selectively disable hypusination of eIF5A are needed to evaluate their therapeutic potential in cancer chemoprevention and treatment.

Barbosa et al. (2016) have applied genetic analysis using yeast model to confirm that translation elongation factor eIF5A is conserved through evolution and is necessary to rescue the ribosome during translation elongation of polyproline-containing proteins. In this original manuscript, the authors present investigations on the structural requirements of eIF5A for binding to the 80S ribosome. The putative translation initiation factor 5A (eIF5A) is highly conserved protein from archaea to mammals and, thus far, is the only cellular factor which is posttranslationally hypusinated. Although the site of eIF5A binding to the ribosome is known, no systematic analysis has been performed so far to determine the important residues on the surface of eIF5A required for ribosome binding. This study used clustered charged-to-alanine mutagenesis and structural modeling to address this question, and generated four new mutants of yeast eIF5A.

Chaturvedi et al. (2012) who previously reported the importance of utilizing L-arginine as a host response to the gastric pathogen *Helicobacter pylori*, now show that generation of antimicrobial nitric oxide (NO) by inducible NO

synthase (iNOS), which is dependent on L-arginine availability, is limited by competition with arginase II activity (Lewis et al. 2011) and upregulation of ODC. Spermine blocks L-arginine uptake into macrophages (Chaturvedi et al. 2010) and SMO mediates DNA damage in gastric epithelial cells. Three articles on this topic are included in this issue. The study by Hardbower et al. (2016) is focused on a novel role for arginase 2 (ARG2) in regulating the macrophage response and polyamine metabolism during infection with the gastric pathogen, *H. pylori*. The authors have shown that loss of ARG2 leads to increased histologic gastritis in mice infected with *H. pylori*, and hypothesized that iNOS2 may regulate this response. Now authors report that the immune response to *H. pylori* in Arg2$^{-/-}$mice is not regulated by iNOS2, but instead, loss of ARG2 leads to enhanced M1 macrophage activation, contributing to increased gastritis, reduced *H. pylori* bacterial load and upregulation of polyamine synthesis and catabolism—these may in turn affect disease pathogenesis. Thus, ARG2 has an essential role in pro-inflammatory macrophage activation and function, a previously unsuspected role in regulating polyamine metabolism.

Another interesting role of Arg is described by Lenis et al. (2016). Embryonic survival requires histotrophic nutrition, including molecules secreted or transported into the uterine lumen by uterine epithelia. L-Arg is a common substrate for synthesis of NO, agmatine and polyamines, as well as ornithine, proline, glutamate, creatinine and urea. Agmatine (Agm), a product of Arg decarboxylation, is a substrate for agmatinase for the synthesis of putrescine and other polyamines by sheep conceptuses. The study compared effects of Arg and Agm on the behavior of ovine trophectoderm (oTr1) cells cultured in vitro in regard to proliferation, migration, and adhesion; in addition, this study is related to the secretion of interferon tau, the pregnancy recognition signal in ruminants, as well as expression of genes involved in Arg and Agm transport, and polyamines essential for conceptus development. Results of this study indicate roles for Arg and Agm in the regulation of basic amino acids transport, polyamine synthesis, and catecholamines secretion by oTr1 cells.

Li et al. (2016) describe studies to identify the metabolic source(s) of ornithine for polyamine synthesis and proliferation of endothelial cells (EC), using *N*ω-hydroxy-nor-L-Arg (Nor-NOHA, a specific inhibitor of arginase) and gabaculine (an inhibitor of ornithine aminotransferase (OAT). It was revealed that OAT is quantitatively an important source of ornithine than arginase in these cells and inhibition of both arginase and OAT completely deplete intracellular ornithine, in addition to the depletion of intracellular putrescine and spermidine in EC. Interestingly, neither ornithine was found synthesized via arginine:glycine amidinotransferase in EC, nor putrescine from Arg via ADC and agmatinase. In fact, these studies suggest that it is cytosolic ornithine which is more efficiently used for polyamine synthesis than the mitochondrial ornithine, indicating compartmentalization of this pathway in EC.

Antizymes and antizyme inhibitors (AZI/AZIN) are other key regulators of polyamine levels by affecting ODC and polyamine uptake. AZIs interfere with antizyme functions, leading to increased polyamine biosynthesis, increased cell proliferation, as well as enhanced cell transformation and tumorigenesis (Keren-Paz et al. 2006; Mangold 2006). Murakami et al. (2014) provide evidence that multiple forms of mouse AZIN1 mRNA are differentially regulated by polyamines. They found various Azin1 transcripts are formed by alternative splicing or alternative transcription initiation in mice. Polyamines regulate Azin1 expression at the transcription initiation and selection of splicing acceptor sites for the exon 7, both of which may affect the mRNA level of the active AZIN1 protein.

Further, a novel antizyme inhibitor 2 (AZIN2) has been described by Acosta-Andrade et al. (2016). AZIN2 is present in a limited number of tissues and cell types with a prominent secretory activity, but its physiological role remains unclear. This study used bone marrow derived mast cells (BMMCs) from *Azin2* hypomorphic mice to show that AZIN2 has a role in the biosynthesis of serotonin and histamine in mast cells.

An approach to inhibit cancer cell growth has been the use of polyamines as DNA transporter for gene therapy (Vijayanathan et al. 2014). Because polyamines are positively charged under physiologic ionic and pH conditions, they can interact with negatively charged macromolecules such as DNA and RNA (Thomas and Thomas 2001). Polyamine-DNA interaction stabilizes the double stranded DNA as well as unusual structures, including left-handed Z-DNA, triplex DNA, quadruplex DNA and liquid crystalline DNA. Polyamine-mediated DNA condensation to nanoparticles of approximately 100 nm diameter has application in the development of gene delivery vehicles. Thomas et al. (2016a) have reviewed this area of biotechnological intervention.

Rojano et al. (2016) present a bioinformatic analysis of non-coding genetic variation in histamine receptor genes and mining data to discover functional regulatory elements to prioritise variants for further studies. The importance of the non-coding genome has become clear and variants in these regions have been associated with a range of diseases. These authors have combined data from various sources including genome annotation projects, conservation and eQTL data, to identify SNPs in transcription factor binding sites and enhancer regions that affect gene expression. Thus, findings presented here provide a catalogue of potentially functional SNPs in these genes.

On a different course, Sakanaka et al. (2016) summarize a growing body of evidence to support health promoting effects of intestinal polyamines. Although intestinal polyamines are mainly synthesized by intestinal microbiota species, current knowledge on this is limited. These authors elucidate the function of carboxyspermidine decarboxylase of *B. thetaiotaomicron*, a predominant intestinal bacterial species in human, at genetic and biochemical levels.

The second part of this issue is dedicated to novel aspects of TGases related to their applications in material science and their potential as biotechnological tools. TGases were first described as enzymes that facilitate the formation of *N*-ε (γ-glutamyl)lysine isopeptide bond between endo-γ-glutaminyl and endo-ε-lysyl protein residues, or the covalent incorporation of diamines and polyamines in proteins (Folk 1983). Scarnato et al. (2016) focus on the effects of TGases and sourdough on protein profiles and volatile molecules of gluten free dough. The results show that the combined use of cross-linking TGases and a selected microbial consortium of *Lactobacillus sanfranciscensis* and *Candida milleri* were synergistic, showing the capacity to modify gluten free flour proteins and to improve protein network formation. These studies also throw light on the composition of volatile molecules important for sensory properties of the dough as well as for the shelf-life of the final product.

Aloisi et al. (2016) show that plant TGases sequenced so far have little sequence homology with the best-known animal enzymes, except for the catalytic triad. Further, it is revealed that plant TGase activity is present in different organs and sub-cellular compartments, likely with different functions. Also in this issue, the features and roles of TGase in the better studied experimental plant models are described. These include (1) TGase affect organization and dynamic of cytoskeleton proteins in germinating pollen tube; (2) light regulated TGase in chloroplasts stabilizes the photosynthetic complexes and affects photoprotection mechanisms; (3) role of TGase in programmed cell death pathways during self-incompatible (SI) response and leaf senescence.

In conclusion, the carefully selected contents of this special issue reflect the present knowledge and future perspectives of the physiological, biochemical and therapeutic actions of both polyamines and transglutaminases.

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Enrico and Enrica Sovena Foundation was constituted in 1990 and legally recognized as an "Ente Morale" (non profit organization) at the end of 1993. The Foundation started its activities only in 1994 and, from the beginning, the President was Professor Dario Piccinelli. He developed his studies in pharmacology and was full professor and head of the Pharmacology, Pharmacognosy Institute at "La Sapienza" University of Rome, the largest University in Europe established on April 20, 1303 by the bull of the Pope Bonifacio VIII. Professor Piccinelli passed way on March 2016. With this special issue, we wish to express our gratitude to Professor Dario Piccinelli and to Enrico and Enrica Sovena Foundation for the effort in economically supporting young scientists, worthy students, graduates in medicine and surgery, and other medical disciplines in their research fields. We all will miss Dario, but hope to maintain his precious work that will remain an important guide for the many colleagues, and for the new President Professor Gabriela Mazzanti, who shared with him the deep interest for active research and following the aims of the Foundation, mainly for championing the new generation to get involved with enthusiasm and productivity in the medical field.

Appendix: Contributors

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