

Chapter 2

Seed Proteomics: An Overview

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Abstract Seed is vital for propagation of spermatophytes in biome and as food source for inhabitants of the earth. Studies on seed proteins provide platform for new avenues to explore molecular networks and pathways governing seed filling, maturation, germination, and seedling formation. Protein expression changes of three genetically different sub-regions of angiosperm seeds are reflected in ordered chain of biological events represented from family differences in different taxa. Different families of angiosperm show divergence of seed protein evolution and thus provide insights into seed structure and function. A gamut of information is available on seed proteomic datasets from approximately 3500 proteins that impinge on protein function in diverse plant families. The functional modularity of seed proteins were compared amongst species that span from dicot to monocot and diploid to polyploid. Transitions of protein complement revealed difference between dormancy and germination towards understanding biological check point at translational level. Goal of this chapter is to critically review data available till date on seed proteomic studies and identify family and cross genera knowledge gaps. The information thus obtained would unravel new components and an unparallel understanding of the molecular processes underlying translational and post-translational variations under different conditions that involves histodifferentiation and organogenesis of the seed.

Keywords Crop plant · Seed proteomics · Seed · Dormancy · Germination

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2.1 Introduction

Evolution of angiosperm has facilitated the transitional dominance of seed producing plants into a terrestrial environment. According to the fossil records their first appearance dated back to 365 million years ago when majority of landscape was successfully and naturally selected for angiosperm survival [1]. The physiological and genetic control of seed is in part responsible for dispersal mechanism which proved to be a spectacular phenomenon responsible for prevalence of seed plants on livable planet, the earth. True seed is fertilized mature ovule possessing an embryonic plant, stored food, and a protective coat. A major factor during angiosperm embryogenesis is the switch from the radial symmetry of the globular embryo to the bilateral symmetry followed by differentiation of cotyledons and embryonal axis [2]. Seed development and germination is a continuous and fine-tuned process with natural circumscription engrossing three phases of embryogenesis recognized as rapid cell division, deposition of reserves, and desiccation. Commonly, mature seeds are classified as albuminous and ex-albuminous depending on the presence or absence of endosperm. Earlier in 1946, Martin studied the internal morphology of seeds belonging to 1287 genera of angiosperm and classified them based on size of the embryo in relation to endosperm and differences in the size, shape, and position of embryo in the seed. The developmental program of monocot and dicot embryogenesis is different but a highly ordered phenomenon. In addition, regulatory pathways, dissection of complex traits, and developmental reprogramming reflects the fundamental differences of the molecular biology of two taxas.

To build new perspectives approach for the physiological and biochemical factors controlling seed traits in two taxas, omics studies of seed development were performed since year 2000 exploiting the availability of genome sequence and related resources [3]. Further, advancement in high-throughput proteome analysis using gel and non-gel based approaches provided detailed protein profile at different developmental stages which might help to elucidate the regulatory network of embryogenesis related proteins. Currently, proteomics is playing an important role in: (i) understanding plant biology, (ii) developing plant biomarkers for human health and food security and (iii) food analysis and bio-safety issues [4]. Seed proteomics research has largely focused on agriculturally important crop plants. Typically, these seeds are obtained from a commercial source. It is reasonable to assume that they are genetically uniform thus it is not biased by the contributions of a “contaminating” proteome. The strategy of using combinatorial-ligand random peptide beads appears to have substantial potential to deplete the supra-abundant SSP from input samples [5]. In addition, proteomic approaches also dissected double fertilization reprogramming at translational and post-translational level in two different clades of angiosperm. This review aims to compile proteomic analyses performed until now to understand evolution of monocot and dicot seed protein patterning, analyzing protein profile of inaccessible regions of seeds, regulatory networks of underlying mechanism of embryogenesis and filling to modulate and outline the strategies to identify candidate seed proteins controlling the regulatory switches.

2.2 Perspect of Seed Biology

Seed is a multifaceted organ which develops from fertilized ovule and is important for plant survival, evolution and agricultural production. Strictly defined, seed development is accompanied with many distinct metabolic, cellular and physiological changes including imbibition, respiration, RNA and protein synthesis, enzyme activities within its surviving structures like endosperm, nucellus, cotyledon, teguments and components including funiculus and integument. Seed biology research has been conducted extensively due to its importance in food industry. Seeds of different genera and family of angiosperm have diverse importance being used for oil, spices, fibre, carbohydrate, fat, secondary metabolites and in brewing industry. They ensure the perpetuation of life forms and spread of the species to new areas by means of autochory, anemochory, hydrochory, and zoochory [6]. Distinct physio-chemical properties of diverse angiosperm seeds compelled the researchers to study their molecular entities including protein complement to understand cellular circuitry correlated to the morphological and physiological adaptations from germination to seedling growth.

2.3 Protein Organization of Seed Parts

In angiosperm, the female gametophyte is seated deep in the ovarian cavity far away from stigma where pollen germinates. There is sufficient evidence to suggest that gametophyte specific proteins were present during fertilization phase [7]. Further, the triploid endosperm, the most common nutritive tissue for the developing seed in angiosperm, maintains a critical protein balance with embryo and maternal tissues. Nuclear, helobial and cellular endosperm shows either symmetric or asymmetric growth that is exemplified by differences in protein patterns [8]. He et al. [9] performed comparative proteomic analysis of wheat embryo and endosperm during seed germination. The most abundant proteins both in the embryo and endosperm were found to be seed storage proteins such as legumins, vicilins and albumins. Housekeeping enzymes, actin-binding profilin, defense-related protein kinases, nonspecific lipid transfer protein and proteins involved in general metabolism were also identified. In monocot, morphologically and biochemically distinct outermost layer of endosperm namely aleurone exhibit differences in protein organization and constitutes an important accumulatory reserve tissue. Following a predetermined mode of development, fertilized egg give rise to embryo which show differences in morphology, anatomy and biochemistry both in monocot and dicots, and protein composition is no exception. Proteomic studies had begun to reveal proteins that are necessary for the events such as pattern formation, cell differentiation and organ development. Up till now approximately 1500 diverse proteins are reported to be active in dicot plants and 928 proteins in monocot plants (Table 2.1). Many of these are expressed in specific cell types and regions of seeds. For modular

Table 2.1 A comprehensive list of angiosperm seed proteome study

Clade	Family	Plant	Organ/tissue	References ^a
Dicot	Brassicaceae	Arabidopsis	Whole seed	[25]
			Whole seed	[23]
			Whole seed	[19]
			Whole seed	[21]
			Whole seed	[27]
			Whole seed	[26]
			Whole seed	[22]
			Whole seed	[17]
			Whole seed	[18]
		Camelina	Whole seed	[48]
		Castor bean	Nucellus	[45]
			Whole seed	[44]
			Whole seed	[42]
			Whole seed	[46]
		Mustard	Whole seed	[30]
			Whole seed	[49]
		Oilseed rape	Embryo	[35]
		Rapeseed	Cotyledon	[31]
			Endosperm	[34]
			Whole seed	[28]
			Whole seed	[33]
			Whole seed	[32]
			Whole seed	[47]
			Whole seed	[29]
		Rapeseed, arabidopsis	Whole seed	[43]
	Euphorbiaceae	Jatropha	Embryo and endosperm	[36]
			Embryo, endosperm	[37]
			Endosperm	[41]
			Inner integument	[39]
			Whole seed	[40]
			Whole seed	[38]
	Leguminosae	Chickpea	Whole seed	[80]
		Common bean	Whole seed	[78]
		Lentil	Whole seed	[79]
			Cotyledons, hypocotyl	[73]
			Endosperm,	[74]
			Whole seed	[71]
			Whole seed	[72]

(continued)

Table 2.1 (continued)

Clade	Family	Plant	Organ/tissue	References ^a
		Medicago	Whole seed	[68]
			Whole seed	[66]
			Whole seed	[69]
		Medicago, black bean	Whole seed	[70]
		Mungbean	Whole seed	[81]
		Pea	Embryonic axis	[57]
			Whole seed	[75]
			Whole seed	[76]
		Peanut	Whole seed	[82]
			Whole seed and testa	[83]
		Pigeon pea	Whole seed	[84]
		Soybean	Cotyledon	[64]
			Cotyledon	[63]
			Hypocotyl, radicle	[65]
			Seed coat	[13]
			Seed coat	[12]
			Whole seed	[51]
			Whole seed	[52]
			Whole seed	[5]
			Whole seed	[60]
			Whole seed	[61]
			Whole seed	[58]
			Whole seed	[62]
			Whole seed	[59]
			Whole seed	[57]
			Whole seed	[56]
			Whole seed	[55]
			Whole seed	[54]
			Whole seed	[53]
			Whole seed	[59]
	Amaranthaceae	Sugarbeet	Whole seed	[85]
	Anacardiaceae	Cashew	Cotyledon	[86]
	Cucurbitaceae	Melon	Whole seed	[89]
	Oleaceae	Olive	Whole seed	[90]
	Rosaceae	Cherry	Cotyledons, embryos, testae	[88]
	Rubiaceae	Coffee	Embryo	[92]
	Solanaceae	Tomato	Embryo, endosperm	[87]
	Theaceae	Tea	Whole seed	[93]
	Vitaceae	Grape	Endosperm	[91]

(continued)

Table 2.1 (continued)

Clade	Family	Plant	Organ/tissue	References ^a
Monocot	Poaceae	Barley	Aleurone	[150]
			Aleurone, endosperm, embryo, whole Seed	[144]
			Endosperm	[143]
			Whole seed	[154]
			Whole seed	[147]
			Whole seed	[142]
			Whole seed	[145]
			Whole seed	[148]
			Whole seed	[149]
			Whole seed	[146]
			Whole seed	[152]
			Whole seed	[151]
			Whole seed	[153]
		Maize	Embryo	[122]
			Embryo	[135]
			Embryo	[134]
			Embryo	[133]
			Embryo	[132]
			Embryo, endosperm	[139]
			Embryo, endosperm	[140]
			Embryo, endosperm	[102]
			Endosperm	[138]
			Scutellum	[136]
		Rice	Whole seed	[137]
			Embryo	[114]
			Embryo	[115]
			Embryo	[116]
			Embryo	[109]
			Embryo	[108]
			Embryo	[107]
			Embryo	[106]
			Endosperm	[110]
			Endosperm	[111]
			Endosperm	[113]
			Whole seed	[105]
			Whole seed	[104]
			Whole seed	[57]
			Whole seed	[101]
			Whole seed	[100]

(continued)

Table 2.1 (continued)

Clade	Family	Plant	Organ/tissue	References ^a
			Whole seed	[99]
			Whole seed	[97]
			Whole seed	[103]
		Wheat	Amyloplast	[128]
			Amyloplast	[127]
			Endosperm	[125]
			Endosperm	[130]
			Endosperm	[129]
			Endosperm	[126]
			Endosperm	[131]
			Whole seed	[124]
			Whole seed	[119]
			Whole seed	[120]
			Whole seed	[118]
			Whole seed	[117]
			Whole seed, embryo	[123]

^aBased on PubMed search dated November 5, 2015

organization of seeds belonging to different clades of angiosperm, expression of proteins unique to each autonomous region must be regulated at both translational and post-translational level. During development or maturation it is typical to analyze whole seeds as the seed coats and embryonic axes make a relatively small contribution to the total mass [10]. Besides acting as a physical barrier, the seed coat has other multifunctional roles majorly in the metabolic control of seed development and dormancy, disease resistance and in nutrient metabolism from parent plant [11, 12]. In case of soybean seed coat, shotgun proteomic approach was used to identify 1372 seed coat proteins majorly involved in primary and secondary metabolism, cellular structure, stress responses, nucleic acid metabolism, protein synthesis, folding and targeting, hormone synthesis, signaling, and biogenesis of seed storage proteins (SSPs) [13].

2.4 Prototype Extraction of Seed Proteins

Being a sink organ, seeds of angiosperm have reserves of carbohydrate, fat, protein, oil, secondary metabolites, organic acids and cyclic compounds, which make the extraction of protein a daunting task. Total protein content in different angiosperm families vary and show differences in stability, activity and selectivity. Seeds were subjected to protein profiling much before the concept of proteome had emerged. Most of the seed proteomic studies have been carried out using differential

biochemical extraction method for fractionation followed by resolution utilizing two dimensional gel electrophoresis (2-DE). Study of seed proteins dates back for over 270 years with the isolation of wheat gluten in 1745 by Beccari. Thereafter, systematic studies on seed proteins were carried out by Osborne in 1924 who classified seed proteins based on their solubility and extraction in a series of solvent, for example, water soluble (albumins), dilute saline soluble (globulins), alcohol soluble (prolamins) and dilute acid or alkali soluble (glutelins). More systematic seed protein extraction was carried out in 1965 that focused on the salt soluble seed proteins from pumpkin by extraction through column gradient [14]. Subsequently, seed protein extraction was performed utilizing weak buffer at neutral pH and low ionic strength by Kriz [15]. These conditions were found to enrich low abundant proteins, which are otherwise masked by the presence of high abundant storage proteins. Furthermore, Kreis et al. in 1985 reported extraction of proteins in only deionised water. Improvement in the fractionation procedure underlined of seed sub-regions namely, embryo, endosperm, aleurone, scutellum, cotyledons involved dedicated extraction methods like tris-phenol, acid or alkaline extraction, two-phase separation and urea solubilization [10]. Organellar proteomes from seeds are very less studied due to unavailability of sufficient material.

2.5 Seed Proteomes: A Composite Insight

2.5.1 *Seed Proteomics of Angiosperm*

Evolutionary relationship between monocot and dicot seeds is an outcome of protein diversity that could be interpreted from the translational landscape obtained from proteomic approaches. Protein signatures in seeds of diverse taxa provide an integrated view based on molecular characteristics. Sub-regions of seed reveal similarities and differences in biological processes among different taxa depicting common themes in diverse genera. Unraveling the molecular basis of embryogenesis in angiosperm at protein level has laid a foundation for the rational improvement of agricultural production. Seed sub-regions have features that appear to be common in angiosperm, including conserved systems for deploying developmentally related proteins and storage strategies [16]. Studies on the mechanism of embryogenesis have revealed how different sub-regions of seed have mimicked fundamental strategies for protein communication in angiosperms. Buildup of precursor metabolites, rapid endoreduplication, growth of pod, testa and endosperm leading to increase in storage protein synthesis and cell division suggests that conserved developmental events exist among angiosperm, although difference lie at molecular, anatomical and physiological levels. These findings highlight the utility of the cross-taxa comparison of seed proteomes for rigorous dissection of fundamental components of seed developmental machinery (Fig. 2.1).

2.6 Assessment of Seed Sub-region Proteomes of Eudicot

2.6.1 Brassicaceae Seed Proteomics: Sub-region Protein Functions

In total, twenty-eight seed proteomes have been reported till date encompassing twenty-four whole seed, one each on nucellus, embryo, endosperm and cotyledon (Table 2.1). Brassicaceae is one of the most assorted angiosperm families whose seeds have varied economic importance. The relationship between most of the allied genera is of importance to understand regulatory responses of seed development among family members (Fig. 2.1).

The seed proteome collinearity of several species of Brassicaceae was compared and three major conclusions emerged. First, more than 2000 proteins, about 800 phosphoproteins, and 200 phosphopeptides were identified in Arabidopsis whole seed proteome pointing towards metabolite fluxes restructuring, polar transport of hormones, proteins related to growth and development, desiccation tolerance, germination, dormancy release, vigor alteration and responses to environmental factors. These

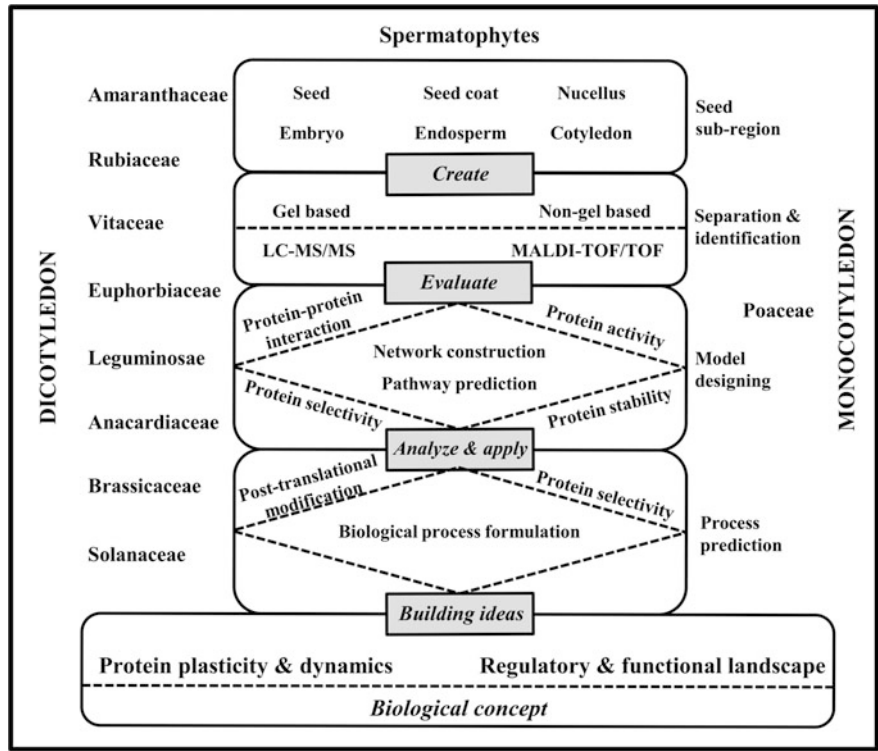


Fig. 2.1 Schematic representation of family based seed proteomics

reports greatly expands knowledge about Brassicaceae seed biology [17–27]. Secondly, around 700 proteins from whole seed, 930 proteins from endosperm and 37 proteins from cotyledons were identified from *Brassica napus* [28–33]. These studies depicted that proteins involved in genetic information processing, carbohydrate metabolism, environmental information processing, energy metabolism, cellular processes and amino acid metabolism were predominant. In endosperm proteome, proteins involved in sugar conversion and recycling, ascorbate metabolism, amino acid biosynthesis and redox balancing were detected confirming the fact that metabolite reallocation and reformation is the major functionality of endosperm during development [34]. Implications of the seed filling process and the function of the embryo were also elaborated in one of the study, where proteins involved in organogenesis, embryogenesis and development were identified [35]. Furthermore, in another study on *Brassica campestris* seed development led to the identification of 260 proteins involved in oxidation/detoxification, energy, defense, transcription, protein synthesis, transport, cell structure, signal transduction, secondary metabolism, transposition, DNA repair and storage [30]. Altogether, these studies revealed conserved protein complexes, processes and functionality amongst the studied species. Nonetheless, diversity lies in signaling and developmental strategies displaying morphological, anatomical, physiological and biochemical variations.

2.6.2 *Euphorbiaceae* Seed Proteomics: Shared and Distinct Proteins

The *modus operandi* in investigating the seed sub-region proteomes of Euphorbiaceae species was the availability of relevant proteomic studies conducted till date. The outcome suggested that 187 whole seed proteins, approximately 5000 endosperm associated proteins, 28 embryo specific and about 3000 integument derived proteins were detected using gel and non-gel based approaches from *Jatropha curcas* (Table 2.1) [36–41]. Data showed that majority of proteins were unique while some housekeeping proteins were common to specific sub-regions. The identified proteins revealed the predominance of protein inhibitor, metabolism, ROS regulated, transport, development and protein degradation related proteins. The creation of prognostic protein modules were used to identify specific regulators operating in the developmental circuitry responsible for coordinating biological processes. Seeds of *Jatropha* are prospective resource of biodiesel generation. Seed proteomic studies would laid a foundation to understand basic information on the biosynthetic pathways associated with synthesis of toxic diterpenes, fatty acids and triacylglycerols and deposition of storage proteins during seed development. These studies provide an important glimpse into the enzymatic machinery devoted to the production of carbon (C) and nitrogen (N) sources to sustain seed development and quality. Another family member, *Ricinus communis* was explored for its seed protein dynamics wherein around 2700 proteins from whole seed and 766 proteins

from nucellus were identified those involved mainly in fatty acid and amino acid metabolism [35, 42–49].

2.6.3 Leguminosae Seed Proteomics: Common and Contrasting Facets

Renovating Leguminosae family for seed proteomic studies proved to be a descriptive and distinct approach for many agriculturally important legumes including pea, Medicago, soybean, mungbean, peanut, Lens, chickpea, common bean and Lotus. Till date, thirty nine seed proteomic studies comprising of whole seed, cotyledons, embryonic axis and endosperm were conducted on legume species to shed light on the mechanism that regulates seed development, morphogenesis and embryogenesis (Table 2.1). Proteomic studies on soybean seed identified 2472 whole seed proteins involved in nitrogen, carbon and lipid metabolism [50–62]. Also, 472 seed coat proteins identified from soybean showed that cell wall associated bioenergetic pathways were integrated with carbon anabolism and catabolism of fatty acids which might contribute to seed coat formation [12, 13]. Further, 328 cotyledon proteins were identified from different cultivars of soybean those associated with oxidative modification of distinct seed-stored mRNAs having role in oxidative phosphorylation, ribosome biogenesis and nutrient reservoir suggesting the significance of post-transcriptional repression of these biological processes regulating seed dormancy [63–65]. Collectively, soybean datasets provide evidence that several regulatory pathways encompassing metabolic, signal transduction and transport related protein contribute to the seed embryogenesis. Cataloging of seed proteins in another agriculturally important crop of the same family, Medicago revealed an imperative corollary, which shows that overall studies until now have been conducted only on whole seed that led to the identification of 308 proteins [66–70]. To comprehend further, Lotus seed proteomic studies when analysed showed the presence of 1500 proteins and 343 phosphoproteins predominantly involved in metabolism and signal transduction forming a regulatory hub that might be controlled by feedback loops [71–74]. Pea seed proteomics spotlight total repertoire of accumulatory and storage proteins from seed sub-regions. It is apparent that primary metabolism, secondary metabolic processes and ROS associated pathways are activated during seed development [75–77]. Unlike soybean, Medicago, Lotus and pea, seed proteomes in chickpea, common bean, Lens, mungbean and peanut are less studied [78–84]. Metabolic pathways were found to be distinct in subfamilies of legumes. Therefore, decoding seed proteome dynamics in less explored legumes are of utmost importance to understand diversity among the protein complement in this family.

2.6.4 Seed Proteomics of Less Studied Plant Families: A Way Forward

To explore less studied plant families, a defined scheme of seed proteome research were adapted that illustrate some protein variants were functionally and structurally modular and involved in developmental processes. Seed specific proteomes of cashew (Anacardiaceae), sugarbeet (Amaranthaceae), coffee (Rubiaceae), grape (Vitaceae), flax, tomato and potato (Solanaceae), Prunus (Rosaceae) and melon (Cucurbitaceae) involving climacteric and non-climacteric fruits of economic importance have been reported (Table 2.1) [85, 86]. In total seed proteomic studies of climacteric fruit tomato (788 proteins from embryo and endosperm), Prunus (1266 proteins from cotyledon, embryo, and testae), and melon (3 peptides from seed) identified an array of proteins associated with ripening, cell wall strengthening, organ development, storage reserve accumulation and embryogenesis [87–89]. Seed proteins from non-climacteric fruits namely grape (3 proteins from endosperm) and olive (231 proteins from seed) were mostly involved in seed formation and development [90, 91] comprising ascorbate peroxidase, amylase, malate dehydrogenase and triose phosphate isomerase. In contrary, seed proteomic studies from plants used for beverages viz., coffee (10 proteins from embryo) and tea (34 proteins from whole seeds) showed the presence of high levels of ROS related proteins that might alter the redox status and determine seed viability [92, 93]. Till date three reports have been published on flax seed proteomes that identified 1744 proteins involved in reorganization of seed cellular machinery during development promoting primary and secondary metabolites reallocation. In addition, oxidative homeostasis, photosynthesis, fruit quality, embryogenesis and development related proteins were also reported in flax seed proteome (Table 2.1) [94–96].

2.7 Assessment of Monocot Seed Proteomes

2.7.1 Grain Seed Proteomics: Overlapping and Unique Proteins

To address key protein complement of monocot seeds, Poaceae family which represents the most extensively investigated members have been used for assessing grain proteome. Of the total 53 proteomic studies on seed sub-regions, rice and wheat having twenty and seventeen reports formed the predominantly studied members whereas fifteen and eleven reports were from barley and maize, respectively (Table 2.1). When rice seed proteomes were explored, twenty one proteomic studies including whole seed (ten reports), embryo (three reports) and endosperm (eight reports) revealed reorganization of protein pool during various developmental stages. 2186 differentially expressed whole seed proteins were found to be involved in central metabolic or regulatory pathways, including carbohydrate

metabolism (especially cell wall synthesis) and protein synthesis, folding and degradation. These provide proteomic confirmation of the notion that seed formation and development involves diverse but delicately regulated pathways [97–105]. Our inclusive analysis of endosperm (408 proteins) and embryo (1656 proteins) proteomics of rice, a valuable proteomic resource highlight characterization of pathways contributing to organ development and embryogenesis at molecular and biochemical levels [106–116]. Phosphorylation, a well-studied post-translational modification is fundamental in the signal transduction cascades during histodifferentiation and embryogenesis. We observed that till date only one report illustrates the role of phosphorylation in embryo formation in rice. The study identified 168 phosphoproteins elucidating the involvement of biomolecular signaling and hormonal interplay during cell division, differentiation and delineation of rice embryo. A glance at wheat seed proteomics yielded a total of 2327 whole seed proteins from ten separate studies (Table 2.1) [3, 46, 117–124]. Wheat seed sub-region proteome have identified 2789 endosperm specific proteins and 63 embryo related proteins [9, 125–131]. Investigating functional and regulatory context of wheat seed proteome research paved the way to understand sink tissue biology of a polyploid crop. Data showed that differentially expressed proteins from embryo were mainly related to carbohydrate metabolism, amino acid metabolism, nucleic acid metabolism and stress-related proteins; whereas those from the endosperm were mainly involved in protein storage, carbohydrate metabolism, protein inhibitors, stress response, and protein synthesis. Translational changes of wheat whole seed display distinct differentially expressed proteins and their synergistic expression provide a mechanistic basis for the normal germination of wheat seeds. Analysis of maize seed proteomes yielded eleven reports including whole seed (one report), embryo (five reports), embryo and endosperm (three reports), endosperm (one report) and scutellum (one report) till date (Table 2.1) [102, 122, 132–140]. Furthermore, 2809 embryo associated proteins involved in organ development, transportation, amino acid metabolism, defense response, molecular chaperone function, protein synthesis, proteolysis, secondary metabolism and signal transduction have been catalogued using gel and non-gel based proteome analysis. Additionally, 183 endosperm related proteins were identified involved in storage, C and N recycling and biogenesis. Molecular basis of seed development of maize was elucidated from whole seed proteotypes that resolved 4511 proteins. Proteins involved in turgor pressure generation, energy metabolism, secondary metabolism, protein synthesis and oxidative burst were identified in different maize genotypes. Further, fifteen seed proteomic studies of barley has identified 423 proteins from whole seed associated with photosynthesis and energy metabolism, carbohydrate metabolism, protein degradation and defense (Table 2.1). Also, 168 proteins from aleurone, embryo and endosperm belongs to diverse functional categories such as metabolite allocation, carbohydrate metabolism, amino acid metabolism, defense response, protein folding and stabilization and oxidative stress tolerance [141–154]. A vast array of monocot seed proteins add diversity to the seed biological properties leading to exclusivity and specificity in cellular processes. Therefore,

characterization of the seed sub-region proteome holds promise of increasing understanding about the regulation of genes and their function in diverse monocot genera.

2.8 Conclusion

Protein expression of recalcitrant or orthodox seed and climacteric or non-climacteric seed belonging to diverse families of angiosperm revealed that the enzymatic machinery initiate germination during the maturation phase is a common theme. The extent to which the functionally active translational machinery would synthesize protein at different stages of seed development differs between different family, genera and species. Protein content and profiles of the seed tissues (cotyledon, embryonic axis and endosperm) were reasonably diverse in different families. The embryonic axis showed proteins related to cell division, histodifferentiation, organogenesis and embryogenesis. Majority of endosperm related proteins, being classified according to their function into major group primarily involved in macronutrient metabolism, metabolite accumulation and assimilation. Cotyledon showed higher number of metabolic and storage related proteins compared to embryonic axis. The tegument, aleurone, nucellus and scutellum presented the largest number of the transport, signaling and cytoskeleton proteins. Protein patterning is in agreement with the biological role of the tissues. Studies on seed tissue and sub-region proteome confirmed a compartmentalization of biological pathways and a partition of metabolic fluxes between different regions of seed. This partition and compartmentalization uncovered the divergence and particularities of the seeds from different families.

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