1. Introduction

Ginseng (Panax ginseng C.A. Meyer) has been used clinically as a tonic or adaptogenic agent for millennia, and nowadays incorporated in functional foods, cosmetics, and beverages (Mancuso & Santangelo, 2017; Yan et al., 2014). Ginsenosides, almost exclusively be found in Panax plants, exert the major pharmacological activities (Kim, Zhang, & Yang, 2015; Leung & Wong, 2010). Tissue culture, bioconversion, and other synthetic biology techniques have been applied to produce ginsenosides to overcome the resource scarcity and meet the market demand (Bulgakov, Khodakovskaya, Labetskaya, Chernoded, & Zhuravlev, 1998; Palazón et al., 2003). The biosynthetic pathway of ginsenosides and expression pattern of related regulatory genes have been investigated by comparative transcriptome (Cao et al., 2015). Xu et al. (2017) performed whole-genome sequencing of P. ginseng, that facilitates genome-wide investigation and reference genome-based transcriptomic profiling of ginsenoside biosynthesis and regulation.
The phytohormone auxin participates in plant growth, development, and responses to environmental stress through its spatiotemporally regulated local accumulation or depletion (Benková et al., 2003). As defense molecules in plant stress and pathogen interactions, ginseng saponins production in the hairy roots of a Panax hybrid increased approximately 1.7-fold with the addition of auxins (0.5 μmol/L 3-Indole butyric acid and 1.0 μmol/L 1-naphthaleneacetic acid) (Washida, Shimomura, Takido, & Kitanaka, 2004). The activity of 3-hydroxy-3-methylglutaryl coenzyme A reductase (HMGR), the first rate-limiting enzyme in triterpene ginsenoside biosynthesis through the mevalonic acid (MVA) pathway, is modified at the transcriptional and post-translational level, including Pleiotropic Regulatory Locus1 (PRL1) (Tholl & Lee, 2011) and negative regulation by protein phosphatase 2A (PP2A) (Leivar et al., 2011). Since PRL1 functions as a regulator of stress and hormone response and PP2A is indeed involved in abscisic acid and auxin signaling, it is assumed that phytohormone and stress response contribute to regulate the HMGR activity (Ursula et al., 2010; Kim et al., 2015). Furthermore, both MVA and mevalonolactone pathways have been shown to be coordinated by hormone molecules via PRL1 (Flores-Pérez et al., 2010). However, the regulatory mechanism controlling genes/enzymes relative to plant triterpene biosynthesis is still little known.

The multiple functions exerted by auxin are largely dependent on its uneven distribution, which is primarily achieved via the vascular system and polarized auxin transport system (Friml et al., 2003; Tanaka, Dhonukshe, Brewer, & Friml, 2006). As the prominent auxin transporters, the PIN-FORMED (PIN) protein family consists of eight members in Arabidopsis thaliana (Paponosov, Teale, Trebar, Billou, & Palme, 2005), among which PIN1–4 and PIN7 are localized to the plasma membrane and govern directional, cell-to-cell auxin transport (Adamowski & Friml, 2015). In contrast, the endoplasmic reticulum-localized PIN5, 6, and 8, together with members of the PIN-LIKE (PILS) family of auxin efflux transporters, mediate intracellular auxin homeostasis (Barbez et al., 2012; Cazzonelli et al., 2013; Ding et al., 2012; Mravec et al., 2009). PIN genes from rice (Wang et al., 2009), sorghum (Shen et al., 2010), maize (Yue et al., 2015), potato (Efstathios, Bjorn, Marian, Visser, & Kitanaka, 2004). The activity of 3-hydroxy-3-methylglutaryl coenzyme A reductase (HMGR), the first rate-limiting enzyme in triterpene ginsenoside biosynthesis through the mevalonic acid (MVA) pathway, is modified at the transcriptional and post-translational level, including Pleiotropic Regulatory Locus1 (PRL1) (Tholl & Lee, 2011) and negative regulation by protein phosphatase 2A (PP2A) (Leivar et al., 2011). Since PRL1 functions as a regulator of stress and hormone response and PP2A is indeed involved in abscisic acid and auxin signaling, it is assumed that phytohormone and stress response contribute to regulate the HMGR activity (Ursula et al., 2010; Kim et al., 2015). Furthermore, both MVA and mevalonolactone pathways have been shown to be coordinated by hormone molecules via PRL1 (Flores-Pérez et al., 2010). However, the regulatory mechanism controlling genes/enzymes relative to plant triterpene biosynthesis is still little known.

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Multiple protein sequence alignments of PIN or PILS were performed using ClustalW in BioEdit Sequence Alignment Editor and were visualized with the WEBLOGO program (http://weblogo.berkeley.edu) to identify conserved amino acid residues. Full-length protein amino acid sequences were used to construct a phylogenetic tree, by employing Jones, Taylor, and Thornton substitution models with parameters of estimated proportion of invariable sites (I) and estimated g-distribution (G). An unrooted neighboring phylogenetic tree of 111 PIN/PILS proteins was constructed using MEGA 7.0 with the following parameters: pairwise deletion option, 1000 bootstrap replicates, and Poisson correction distance. The display, manipulation, and annotation of phylogenetic trees were performed using the Interactive Tree of Life (iTOL, http://itol.embl.de/).

2.2. Multiple alignment and phylogenetic analyses

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2.3. Sequence features and gene structure

The exon–intron structure of each PIN/PILS gene from P. ginseng (PgPIN/PgPILS) was determined by aligning the full-length cDNA sequence or predicted coding sequence (CDS) with the genomic sequence. Gene sequence was displayed by the cross-platform program TBtools 0.53 (http://cj-chen.github.io/tbtools). Subsequently, the online ExPaSy server (https://www.expasy.org/) was used to predict the theoretical isoelectric point (pI) and molecular weight of PgPIN/PgPILS proteins. Conserved motifs in 17 PgPINs and 11 PgPILSs were identified using MEME (Suite version 4.12.0, http://meme-suite.org/index.html) with previously reported criteria (Chu et al., 2018). Finally, the MAST program was used to search for detected motifs (Bailey & Gribskov, 1998).

The subcellular localization of PgPIN/PgPILS proteins was predicted by WoLFPSORT (Horton et al., 2007) based on the 14 nearest neighbors’ features (https://wolfsort.hgc.jp/). HMMTOP 2.0 (Tusnady and Simon, 2001) (http://www.enzim.hu/hmmtop/html/submit.html) and TMHMM Server 2.0 ( Krogh, Larsson, von Heijne, & Sonnhammer, 2001) (http://www.cbs.dtu.dk/services/TMHMM/) were used to predict the structures of PgPIN/PgPILS proteins, which were visualized by TMProt2D (Spyropoulos, Liakopoulos, Bagos, & Hamodrakas, 2004).

2.4. Digital gene expression analysis

Gene expression patterns across different organs of P. ginseng were analyzed using 11 RNA-Seq datasets from NCBI (SRP066368) (Wang et al., 2015), while nine additional RNA-Seq datasets (PRJNA369187 and PRJNA381509) were utilized to compare different tissues (Zhang et al., 2017). Clean reads were separately aligned to assemble the P. ginseng genome in the orientation mode using TopHat (http://tophat.cbcb.umd.edu/)
(Langmead & Salzberg, 2012). Cuffdiff (http://cufflinks.cbcb.umd.edu/) was used for differential expression analysis (Trapnell et al., 2013), using the fragments per kilobase of exon per million mapped reads (FPKM) method to calculate the expression level for each transcript and identify differentially expressed genes (DEGs) among different samples (Wang, Feng, Wang, Wang, & Zhang, 2010). DEGs were identified based on log2 fold change > 2 and false discovery rate (FDR) < 0.05. C. destructans infection was analyzed in seven datasets from *P. ginseng* fibrous roots, where firstly showed the corresponding symptoms (such as reddish-brown spots) at 0.25, 0.5, 1, 4, 7, and 12 days post incubation (DPI) (Gao et al., 2016). The fibrous roots harvested from the uninfected *P. ginseng* plants served as controls (0 DPI). At each time point, the fibrous roots from 3 to 5 *P. ginseng* plants were rinsed with distilled water and then pooled to address the problems of insufficient sample biomass and lack of biological repetition.

Pearson’s correlation test was performed using the R package “Psy” between ginsenoside content and the FPKM of *PgPIN*/PgPILS genes in the nine tissues of the main roots of *P. ginseng* (Zhang et al., 2017). Co-expression analyses between *PgPIN*/PgPILS genes and triterpene saponin biosynthetic enzyme-encoding genes were performed in all 27 RNA-Seq datasets.

### 2.5. Availability of data and materials

The necessary information of public data used in this study are present within the article. Genome sequencing data of *P. ginseng* are available via NCBI under the project number PRJNA3835956. The latest versions of the genome assemblies and annotation are available through our website at http://ginseng.vicp.io:23488/. The GenBank accession numbers for the *PgPIN*/PgPILS genes sequences reported in this paper is summarized in Table S2.

### 3. Results

#### 3.1. Sequence features and phylogenetic analysis

Seventeen *PgPINs* containing open reading frames (ORFs) were identified, with the ORFs being more similar to those of *P. trichocarpa* than of *A. thaliana*, *M. truncatula*, and *O. sativa* (Fig. 1 and Table 1). *PgPINs* gene length varied from 1,373 bp (*PgPIN5a*) to 5,693 bp (*PgPIN6a*), while the length of cDNAs ranged from 864 bp (*PgPIN5a*) to 3,043 bp (*PgPIN3a*) (Table 2). The molecular weights of the predicted proteins ranged from 32,003.30 Da (*PgPIN5a*) to 71,248.39 Da (*PgPIN3b*), and their isoelectric points were predicted to range from 6.42 (*PgPIN5a*) to 9.73 (*PgPIN8b*) (Table 2). Similarly, 11 *PgPILSs* containing ORFs were also characterized (Fig. 1 and Table 1). The number of *PILS* genes in *P. ginseng* was similar to that of *P. trichocarpa* and *M. truncatula*, but greater than that of *A. thaliana* and *O. sativa* (Fig. 1 and Table 1). *PgPILSs* gene length varied from 1380 bp (*PgPILS2a* and *PgPILS2b*) to 11,577 bp (*PgPILS6b*), while the length of cDNAs ranged from 747 bp (*PgPILS6a*) to 1812 bp (*PgPILS1b*) (Table 2). The molecular weights of the predicted proteins ranged from 27,130.08 Da (*PgPILS6a*) to 50,871.47 Da (*PgPILS5a*), and the isoelectric points were predicted to range from 4.94 (*PgPILS1e*) to 9.75 (*PgPILS1b*) (Table 2).

An unrooted phylogenetic tree was constructed using the PIN and PILS proteins in *P. ginseng*, *M. truncatula*, *O. sativa*, *A. thaliana*, and *P. trichocarpa* for subfamily design (Fig. 1). All *P. ginseng* proteins were named based on their relationship with known *A. thaliana* PINs and PILSs, namely, the cluster of PIN and PILS families from *A. thaliana*. The 111 proteins from the above five plant species can be divided into two separate subtrees, suggesting that PILS and PIN proteins are highly conserved in subfamilies but evolutionarily distinct from each other among angiosperms. It was also found that PINs could be divided into eight groups, PIN1, PIN2, PIN3, PIN5, PIN6, PIN8, PIN9, and PIN10, while PILSs could be divided into four groups, PILS1, PILS2, PILS5, and PILS6. No *P. ginseng* members were found in the PIN9 or PIN10 groups, which may have evolved independently in monocots (Adamowski & Friml, 2015; Balzan, Jhobal, & Carraro, 2014). PIN3 and PIN6 exhibited dicospecificity. Within other groups, PINs from each species clustered separately. PIN1 and PILS1 proteins exhibited more distant evolutionary relationships, suggesting that additional clusters could be obtained. This neighbor-joining tree contained 28 PIN/PILS members identified in *P. ginseng*, which indicated a total of 10 distinct subfamilies (Fig. 1). In addition, PIN1 subfamily members from 1 to 5 were dramatically extended in *P. ginseng* compared to that in *A. thaliana*.

#### 3.2. Gene structure and protein profile analysis

The PIN/PILS genes of *P. ginseng* contained a conserved intron-exon organization in several groups (Fig. 2A). The average exon numbers for *PgPIN2*, *PgPIN5*, *PgPILS2*, *PgPILS3*, and *PgPILS8* were 1, 10, 5.5, 6.3, and 4.5, respectively, with the standard deviation being <1. However, gene structures in the *PgPIN1* and *PgPILS7* groups were more variable. The *PgPIN1* group contained five members with the number of exons varying from 6 to 11, with a standard deviation of up to 2.07. Similarly, the *PgPILS1* group contained five members with the number of exons varying from 4 to 7, with a
Gene and protein features of 28 PgPINs/PgPILSs. A central hydrophilic loop (Figs. 2B and 3). All PgPIN hydrophobic segments located at the N- and C-termini exhibited a highly conserved hydrophobicity profile, with two (Fig. 2A and Table S3).

Kleine-Vehn, 2012; Viaene, Delwiche, Rensing, & Friml, 2013). The proteins, but with high heterogeneity in the central hydrophilic loop (Fig. 3C). Multiple sequence alignment and conserved motifs revealed that the sequences of the N- and C-terminal transmembrane segments were highly conserved in PgPIN and PgPILS proteins. motif 8 was absent in two typical short PIN subfamilies (PgPIN5 and PgPIN8). To understand the molecular function of these proteins in P. ginseng, we analyzed their predicted subcellular localization. Most long PgPIN proteins were predicted to be localized in the plasma membrane, while the majority of the short proteins were predicted to be localized in the vacuoles, plasma membrane, and endoplasmic reticulum (Fig. 3B and Table S5).

3.3. Expression profile of PgPIN/PgPILS genes in various organs and tissues

Upon analyzing the expression levels of the 28 PgPIN/PgPILS genes in samples from 11 different organs of P. ginseng, all genes were detected in at least one organ, and seven transport genes were highly expressed in one organ (Fig. 4C and Table S6). Among these seven genes, the seeds expressed three specific PgPINs (PgPIN1d, PgPIN5a, and PgPIN5b), while the roots expressed two specific PgPINs and one PgPILS that were associated with the growth period (PgPIN2b in 12-year root and PgPIN6b in 25-year root; PgPILS1c in 12-year root). PgPIN1c was highly expressed in the fruit pedicel. Five of the seven specific transport genes were not of the typical long type. The remaining 21 PgPIN/PgPILS genes also exhibited altered expression patterns in different organs. As seen in the heatmap (Fig. 4), all identified PgPIN/PgPILS genes clustered into four distinct groups, while eleven samples clustered into three individual groups with specific expression patterns. A total of 13 PgPIN/PgPILS genes belonging to typical short-type subfamilies (PILS1, PILS5, PILS6, PILS7, and PILN8) were differentially expressed, with high levels in the leaf blade and pedicel. Ten PgPIN/PgPILS genes, including six long and two short genes, were more highly expressed in the roots (Fig. 4), while PgPIN1d and PgPILS6b were more highly expressed in the stem.

To further analyze PgPIN/PgPILS expression in P. ginseng roots, the expression of 28 PgPIN/PgPILS genes was examined in three tissues of the main root: the cortex, periderm, and stele. Based on the expression patterns of the 28 PgPIN/PgPILS genes, nine samples were clustered into three distinct groups (Fig. 4B). Four PgPIN/PgPILS genes, including three seed-specific genes, were not detected in any tissue, and 24 PgPIN/PgPILS genes were detected in at least one tissue. Ten PgPILSs and seven PgPILSs across eight

standard deviation of 1.52. As a major factor affecting gene structure, the variations in total intron length (2925 bp vs. 674 bp) were primarily responsible for the striking differences in gene structure between the largest gene (PgPIN1b, 5472 bp) and the smallest gene (PgPIN1e, 2957 bp) in the PgPIN1 group.

Similar to other plants, the P. ginseng PIN/PILS proteins exhibited a highly conserved hydrophobicity profile, with two hydrophobic segments located at the N- and C-termini being linked with a central hydrophilic loop (Figs. 2B and 3). All PgPIN and PgPILS proteins possess 8–10 transmembrane segments (Fig. 2A and Table S3). P. ginseng PINs and PILSs could be classified as long and short based on the length of the predicted protein and the central hydrophilic loop (Feraru, Vosolsobé, Feraru, Petrášek, & Kleine-Vehn, 2012; Viena, Delwiche, Rensing, & Friml, 2013). The long PINs consisted of 10 members (543–657 aa), including all genes from the PIN1, PIN2, and PIN3 groups; the short PINs and PILS1c genes in samples from 11 different organs of P. ginseng, all genes were detected in at least one organ, and seven transport genes were highly expressed in one organ (Fig. 4C and Table S6). Among these seven genes, the seeds expressed three specific PgPINs (PgPIN1d, PgPIN5a, and PgPIN5b), while the roots expressed two specific PgPINs and one PgPILS that were associated with the growth period (PgPIN2b in 12-year root and PgPIN6b in 25-year root; PgPILS1c in 12-year root). PgPIN1c was highly expressed in the fruit pedicel. Five of the seven specific transport genes were not of the typical long type. The remaining 21 PgPIN/PgPILS genes also exhibited altered expression patterns in different organs. As seen in the heatmap (Fig. 4), all identified PgPIN/PgPILS genes clustered into four distinct groups, while eleven samples clustered into three individual groups with specific expression patterns. A total of 13 PgPIN/PgPILS genes belonging to typical short-type subfamilies (PILS1, PILS5, PILS6, PILS7, and PILN8) were differentially expressed, with high levels in the leaf blade and pedicel. Ten PgPIN/PgPILS genes, including six long and two short genes, were more highly expressed in the roots (Fig. 4), while PgPIN1d and PgPILS6b were more highly expressed in the stem.

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subfamilies were more highly expressed in periderm tissue; 10 of the 17 \textit{PgPIN}/\textit{PgPILS} genes were of the short type. In the stele, three \textit{PgPIN1} and all \textit{PgPILS2} members were more highly expressed, none of which were of the short type. Comparing to stele, both \textit{PgPILSS} members were more highly expressed in the cortex and periderm (Table S6).

3.4. Expression profile of \textit{PgPIN}/\textit{PgPILS} genes after \textit{C. destructans} infection

Given that the phytohormone signaling network coordinates the production of defense-related proteins and secondary metabolites during the plant stress response (Gao et al., 2016; Park, Kim, Mishra, & Bae, 2017; Rahman & Punja, 2005), we performed a dynamic transcriptome analysis following infection with \textit{C. destructans}. Based on their expression patterns at 0, 0.25, 0.5, 1, 4, 7, and 12 DPI, 28 genes were clustered into four distinct groups. At T0 (0 DPI), \textit{PgPIN1a}, \textit{PgPIN5a}, \textit{PgPIN8a}, \textit{PgPILS1a}, and \textit{PgPILS6a} showed significantly higher expression, but their expression decreased quickly after infection with \textit{C. destructans}. \textit{PgPIN2a}, \textit{PgPIN2b}, \textit{PgPIN8b}, \textit{PgPILS2a}, and \textit{PgPILS5a} expression increased dramatically at T1 (0.25 DPI), with \textit{PgPIN2b} and \textit{PgPIN8b} only being expressed temporarily. At T2 (0.5 DPI), the expression levels of six \textit{PIN/PILS} genes, namely, \textit{PgPIN1c}, \textit{PgPIN1d}, \textit{PgPIN3a}, \textit{PgPIN3b},
4.1. Herbgenomics facilitates to uncover the genetic information of herbs

Herbgenomics, which aims to explore the genetics and biology of herbs at the genomic level, is proposed as a global platform to identify the synthetic pathways of bioactive compounds (Chen &
Ginseng is one of the most widely used herbal medicines with increasing market demand, on account of its extraordinary ability to maintain physical vitality and increase resistance to aging. Nowadays ginseng has even been developed into healthy nutrition, dietary supplements, and natural cosmetics (Yun, 2001). Furthermore, methods such as BLAST, sequence alignment, phylogenetic analysis, and domain analysis can facilitate the genome-wide identification of genes related to ginsenoside biosynthesis. In this study, we present the genome-wide identification and transcriptional profiling analysis of PIN/PILS auxin transporter gene families in *P. ginseng*.

### 4.2. Structural features of PgPIN/PgPILS contribute to auxin regulation

To coordinate plant growth, development, and secondary metabolism, multiple auxin transporter proteins, including PIN and PILS, function to maintain the directional flow of auxin within different organs through the formation of a spatial and temporal gradient (Adamowski & Friml, 2015; Barbez et al., 2012). In current study, all PgPIN and PgPILS proteins were predicted to localize to membranes and contain putative auxin efflux transport domains (Muday & Murphy, 2002). *In silico* analysis of protein topology indicated that PILS proteins contain a central hydrophilic loop flanked on each side by five transmembrane domains, similar to PIN proteins (Fig. 3). The high similarity in predicted protein topology of PIN and PILS proteins implies similar biological functions in auxin transport (Barbez et al., 2012). Furthermore, PIN proteins can be grouped into long (PIN1-type) and short (PIN5-type) types based on the length of the central hydrophilic loop (Zazimalova et al., 2010). Canonical PIN1-type auxin efflux transporters localize to the plasma membrane and are rate-limiting transporters mediating cellular auxin efflux (Petrášek et al., 2006), while PIN5-type transporters are localized to the endoplasmic reticulum and manage intracellular auxin distribution and homeostasis (Bosco, Dovzhenko, Liu, Woerner, Rensch, Eismann, Eimer, Hegermann, Paponov, & Ruperti, 2012; Ding et al., 2012; Mravec et al., 2009). In our study, PgPIN1, PgPIN2, and PgPIN3 proteins were found to possess a long hydrophilic loop and were, therefore, assumed to localize to the plasma membrane. PgPIN5, PgPIN8, PgPILS1, PgPILS5, and PgPILS6 were predicted to have a short hydrophilic loop and localize to the endomembranes (primarily the endoplasmic reticulum and vacuoles) (Fig. 3). Barbez et al. (2012) reported that all PILS proteins in *A. thaliana* localize to the endoplasmic reticulum, similar to that in *O. sativa* (Mohanta et al., 2015). The majority of PgPILS localized to the endoplasmic reticulum or vacuoles, which may facilitate auxin accumulation inside the cells (Mohanta et al., 2015).

Liu et al. (2017) analyzed the expression patterns of auxin efflux transporter PIN genes in 4-year-old Jilin ginseng and hypothesized that PgPIN2 and PgPIN6 are likely involved in the development and tropism growth in ginseng roots, while PIN3 may be related to the growth and development of the aerial parts of plants. The organ-specific expression profile of PIN/PILS was also confirmed in current study (Fig. 4).

### 4.3. PgPIN/PgPILS probably involved in ginsenosides biosynthesis pathway

In addition to its role in development, auxin is likely involved in secondary metabolism (Sauerwein, Ishimaru, & Shimomura, 1991; Sauerwein, Yamazaki, & Shimomura, 1991). Ginsenosides show a tissue-specific accumulation and distribution and are even heterogeneously distributed in ginseng roots (Liang et al., 2015; Xu et al., 2017; Zhang et al., 2014). At gene level, multiple genes involved in ginsenoside synthesis are expressed in the phloem and xylem, while ginsenosides are primarily distributed and stored in the periderm (Kim et al., 2015; Zhang et al., 2017). More specifically, we measured the ginsenoside content and gene expression in the periderm, cortex, and stele, which were separated from the ginseng main root (Fig. 4). Ten PgPINs and seven PgPILSs were distributed across eight subfamilies with higher expression levels in periderm tissue, with 10 of the 17 PgPIN/PgPILS being of the short type. This implies the potential role of auxin and its transporters in the distribution and accumulation of ginsenosides (Fig. 4).
The production of ginsenosides and root growth in hairy roots in *Panax* hybrids is promoted by the addition of auxins (Washida et al., 2004); however, little is known about the potential mechanism. As stated earlier, it is possible that HMGR activity is regulated by phytohormones and stress (Fig. 7). Furthermore, both the MVA and MEP pathways are coordinated by hormone molecules via PRL1 (Úrsula et al., 2010). In the current study, we found a significant correlation between *PIN/PILS* expression and ginsenoside content, especially DXS and HMGR. Additionally, triterpene function in plant growth and development, both of which are regulated by auxin (Kim et al., 2015); for example, the allelopathic effect of ginsenosides on growth (Zhang, Lei, Fang, Jia, & Zhang, 2011). Total ginsenosides, panaxadiol ginsenosides, and ginsenosides-Rb group inhibited the seedling growth of *P. quinquefolius* at high concentrations but had stimulatory effects at low concentrations. In contrast, panaxatriol ginsenosides stimulate seedling growth at various concentrations. However, the spatiotemporal distribution of auxin depends on a complex interplay between auxin metabolism and intercellular auxin transport. We just found the correlation between *PIN/PILS* and the ginsenoside biosynthetic pathways in the current study, the underlying mechanism and more details involved in auxin transport and ginsenoside biosynthesis remains to be elucidated (Fig. 7).

5. Conclusion

In summary, a genome-wide search of the *P. ginseng* genome for homologous auxin transporter genes identified 17 *PIN* and 11 *PILS* genes. Sequence analysis, putative motif organization, and subcellular localization indicated potential redundant and complementary biological functions of these *PIN/PILS* genes. Furthermore, organ/tissue expression patterns, together with the significant correlation between *PIN/PILS* expression and ginsenoside content,
Fig. 7. Speculated possible regulatory pathway of ginsenosides biosynthesis by PIN/PILS auxin transporters. Auxin transporter PIN/PILS family proteins, mainly locating on plasma membrane and endoplasmic reticulum membrane, maintain the distribution and homeostasis of auxin. PIN2A, a heterotrimorphic enzyme that binds the N-terminal region of HMGR, is involved in auxin, abscisic acid and ethylene signaling and emerges as a positive and negative multilevel regulator of plant HMGR. Meanwhile, PRL11 functions as a regulator of stress, and hormone response. From the above, it is assumed that phytohormone and stress response contribute to regulate the HMGR activity and subsequent ginsenoside production.

suggested that auxin transporters are potentially involved in the regulation of ginsenoside biosynthesis and its local accumulation. Genome-wide comparative analysis of PIN/PILS genes and their expression profile provide a new perspective to understand the molecular basis and regulatory mechanisms of PIN/PILS in P. ginseng, including their roles in ginsenoside production and stress response.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.chmed.2021.08.001.

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