Online Resource 1 Supplementary figures

Online supplementary material for:

**Identification of molecular processes that differ among Scots pine somatic embryogenic cell lines leading to the development of normal or abnormal cotyledonary embryos**

Tree Genetics and Genomes

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Schematic presentation of the cloning strategy design for the construction of **PaCYP78A7** (a) and **PaTT7** (b) RNAi constructs. Green and Blue arrows represent the two fragments used to produce the RNA hairpin construct. Primer sequences are presented in Table S3.

**Fig. S1** Schematic presentation of the cloning strategy design for the construction of **PaCYP78A7** (a) and **PaTT7** (b) RNAi constructs. Green and Blue arrows represent the two fragments used to produce the RNA hairpin construct. Primer sequences are presented in Table S3.
Fig. S2 Quantitative real-time PCR analysis of the relative transcript level of the target gene in transgenic lines of Norway spruce. The transcript level of EXPB1 in 35S:EXPB1 sublines, RIC3 in 35S:RIC3 sublines, SERK1 in 35S:SERK1 sublines, CYP78A7 in 35S:CYP78A7i sublines, and TT7 in 35S:TT7i sublines were analysed. Each analysis included expression of the transcripts in U-control and T-control (35S:GUS) lines. The transcript level was analysed in proliferating embryogenic tissue at stage 1, except for 35S:TT7i sublines where embryos at stage 4 were analysed owing to the low expression at stage 1. The transcripts levels are relative to the transcript level of each gene in the U-control and normalized against one reference gene (EF1). The presented data are means (± SD) of three biological replicates analyzed with two technical replicates each. Different letters indicate significant differences in the relative transcript abundance among sublines (Student’s t-test, P ≤ 0.05).
Fig. S3 Heat map comparing the transcript accumulation pattern of 18 selected genes during somatic embryo development in the cell line 12:12 (transcript name in black), giving rise to normal embryos and the cell line 3:10 (transcript name in blue), giving rise to abnormal embryos, based on data presented in Fig. 1. Relative expression values at six consecutive stages (Table S4) were calculated by the Livak method \((2^{-\Delta\Delta Ct})\) and normalized against two reference genes (Fig 1). The average of the relative expression values of three replicates was used to generated a heat map using the Clustvis webtool (https://biit.cs.ut.ee/clustvis/). Transcript abundance is represented by a scale of color intensity, where red denotes high expression and blue, low expression.
Fig. S4 Heat map showing the transcript accumulation pattern of 18 selected genes during somatic embryo development in Norway spruce based on data presented in Fig. 2. The relative transcript level at eight consecutive stages (Table S4) were calculated by the Livak method ($2^{\Delta\Delta Ct}$) and normalized against two reference genes (Fig. 2). The average expression level of three biological replicates was used to generated a heat map using the Clustvis webtool (https://biit.cs.ut.ee/clustvis/). Transcript abundance is represented by a scale of color intensity, where red denotes high expression and blue, low expression.
**Fig. S5** Differentiation of somatic embryos after one week on maturation medium. (a) T-control. Note the few well developed late embryos. (b) Subline 35S:SERK1.1-1. Note the high frequency of early embryos with irregular embryonal masses. Bars, 100µm