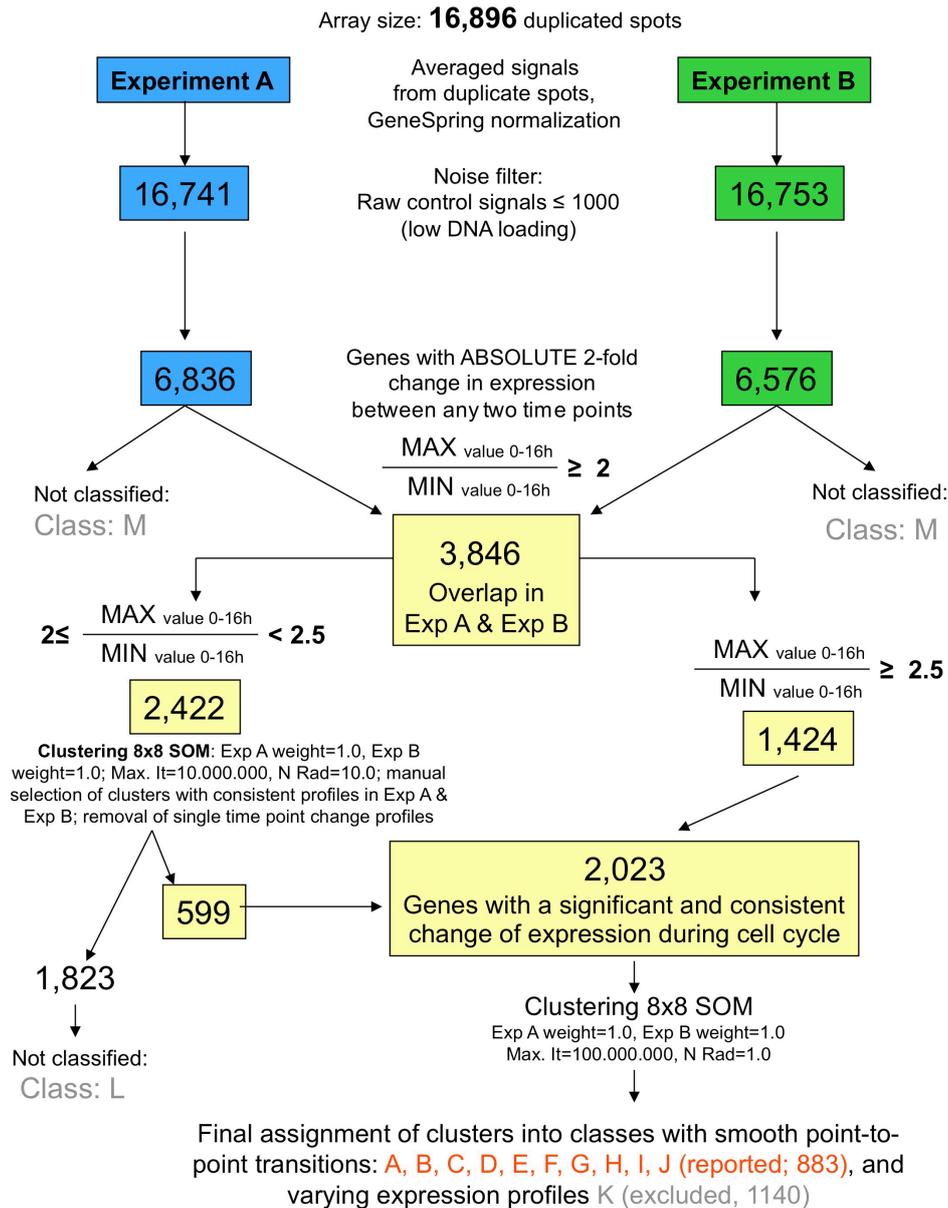
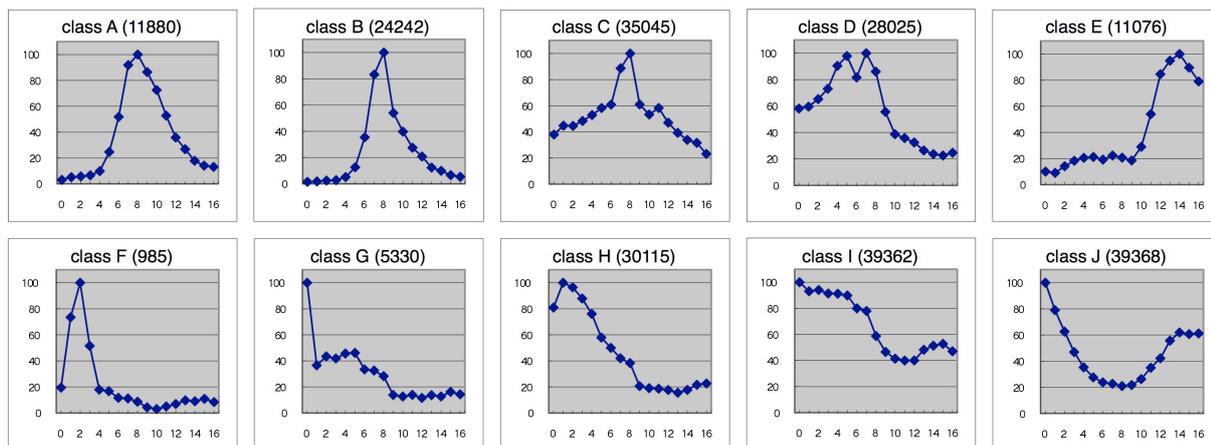


**Supplemental Figure S1.** Cell cycle synchronization of BY2 cells. Synchronization of BY2 cells was performed twice independently in experiments A and B for microarray analyses. In each experiment, the cells were synchronized by aphidicolin treatment and sampled at the indicated times after aphidicolin removal. A, Change in mitotic index during synchronous culture. B, RNA gel blot analysis of *CYCB1;3* and *PCNA* mRNAs. Total RNA was extracted from cells at the indicated times after aphidicolin removal. The RNA blots were hybridized with *CYCB1;3* and *PCNA* probes. C, Flow cytometric analysis of synchronized BY2 cells. The nuclei were extracted from cells sampled at indicated times in synchronous cultures. After staining with DAPI, the fluorescence intensity of the nuclei was measured with a ploidy analyzer. Only the results from experiment A are shown; similar results were obtained in experiment B (data not shown).

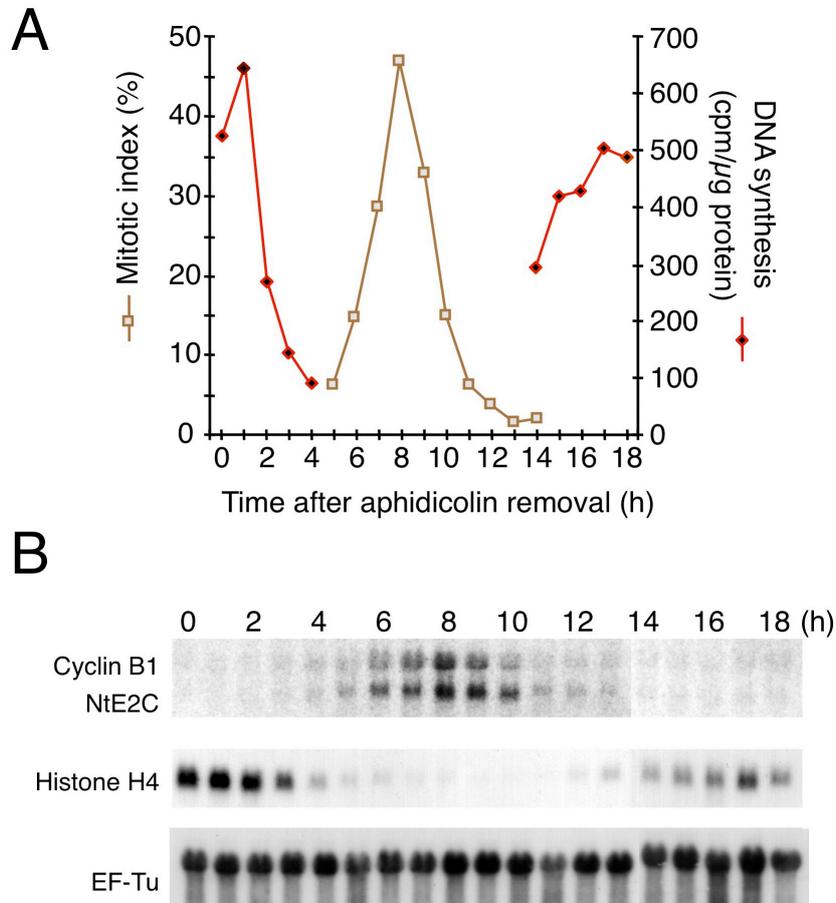
## Processing of microarray data



**Supplemental Figure S2.** Flow chart showing microarray data processing of cell cycle-regulated transcripts. Two biological replicates of the experiment were used to classify the genes based on their expression at 9 time points after release from the aphidicolin block. The values from two technical replicate spots on the microarray were averaged and normalized as described in ‘Materials and Methods’. Spots with arbitrarily selected low Cy-3 signal value (raw signal value  $\leq 1000$ ), indicating small amount of DNA spotted on the chip, were removed by a noise filter. Each spot was evaluated for a 2-fold absolute change between any of the time points in the kinetic experiment, and 3,846 spot overlap between experiments A and B was then used for Self-Organizing-Maps (SOM) clustering to identify genes with similarly changing transcript profiles during the cell cycle. Clusters with consistent profiles were manually selected from 64 SOM clusters generated by the GeneSpring program using the following criteria: (1) clusters showing erratic behaviour or inconsistent profiles in two experiments A and B were excluded, and (2) clusters with similar profiles were pooled to create 10 putative cell cycle groups named A-J containing 883 microarray spots. An annotated list of the 883 spots is shown in Supplemental Table S4, and the raw normalized data for all spots in the two experiments are shown in Supplemental Table S3.



**Supplemental Figure S3.** Real-time RT-PCR analysis of the representative transcript in each of the 10 cell cycle-regulated classes. In each graph (A-J), time points in the synchronous cultures are indicated in h on the  $x$ - axes, and the relative transcript levels are shown on the  $y$ -axes. The results of the real-time PCR are shown after normalization to the expression of *CDKA* mRNA. The gene ID of the transcript analyzed in each class is given in parenthesis (gene ID/GenBank accession number: 11880/BP527114, 24242/BP531347, 35045/BP535211, 28025/BP532602, 11076/BP526359, 985/BP129153, 5330/BP133099, 30115/BP533426, 39362/D89636, 39368/AB025029).



**Supplemental Figure S4.** Accumulation patterns of *NtE2C* and cyclin B1 mRNA in BY2 cells during the cell cycle. A, After aphidicolin removal,  $^3\text{H}$ -deoxythymidine 5'-triphosphate incorporation (solid diamonds) and mitotic index determination (open squares) were used to monitor the progression throughout the cell cycle. B, Gel blot analysis of RNAs extracted from cells at different time points of the synchronization experiment. Twenty micrograms of total RNA was separated by electrophoresis on an agarose-formaldehyde gel, transferred to a nylon membrane, and successively hybridized with *NtE2C* and cyclin B1, and histone H4 and EF-Tu probes.