## lannone et al. Supplementary Figures.



## Figure S1.

**Figure S1. Semi-quantitative RT-PCR validation of alternative splicing changes predicted by MISO analysis of RNA-Seq data.** Histograms represent log2 fold change of % exon inclusion in cells treated with 10nM of R5020 for 6h versus untreated cells, calculated using MISO or using semiquantitative PCR followed by capillary electrophoresis. Error bars represent standard deviation. We considered an event as validated when the direction of the changes detected by the two methods concur. Using this criteria, 5 (*CLUHP3*, *DMTF1*, *DTNBP1*, *FAM126* and *HSF1*) out of 7 events analyzed were validated.

Figure S2.



Position relative to 3' splice site (bp)

**Figure S2.** General nucleosome density profiles in exons, grouped according to their Percent Spliced In (PSI) index, and flanking sequences in control (T0) and hormone-stimulated (T60) T47D cells. Lower panels: separate profiles for PSI  $\ge$  95% (left) or PSI  $\le$  5%(right).

Figure S3.



Figure S3. Meta-profile of the ChIP-Seq signal of total RNA Polymerase II in untreated (blue line) and R5020-treated cells (red line) relative to the transcription initiation site. The figure shows accumulation of RNA pol II on Transcription Start Sites and reduced signal over the gene body. The two plots overlap extensively.

## Figure S4.



Figure S4. RNA polymerase II density from ChIP-Seq data relative to exon/intron structure. Boxplots represent the density of deep sequencing reads corresponding to alternative exons and 750 upstream intron nucleotides, normalized to local average in mock-treated with ethanol (blue) or with R5020 for 60 minutes (red). Density maps correspond to input DNA (A,D) or RNA pol II ChIP-Seq signals corresponding to R5020-included (B), R5020–skipped (E) or their corresponding downstream exons (C,F). P-values calculated by paired two-sample t-test are indicated by \* (0.05>p>0.01), \*\* (0.01>p).

Figure S5.



**Figure S5.** General profile of nucleosome density in annotated alternative exons (the same as used for clustering in figure 4A) and flanking sequences in control and hormone-stimulated T47D cells.