Suppl. Figure 4B  Huettel et al.
Suppl. Fig. 4. (A) The intergenic region on chromosome 2 giving rise to the IG1 transcript that is induced in the drd1 mutant. Top: The IG1 transcript detected by cDNA-AFLP is depicted as a black bar with the orientation indicated by the arrow. This fragment overlaps with a full length cDNA (light gray bar) from the Riken collection (RAFL22-40-J02; http://rarge.gsc.riken.jp/cdna/cdna.pl) and a transcript detected primarily in callus in a whole genome array (dark gray bar) (http://signal.salk.edu/cgi-bin/atta). Sense and antisense short RNAs, detected by MPSS (Lu et al., 2005) are indicated by vertical arrowheads on the top and bottom of the double dotted line (dark, unique; white, multiple matches). The short RNA-encoding sequence is unannotated and does not show significant similarity to any known transposon or gene. An ~ 400 bp region upstream of the IG1 transcript contains 15 CG dinucleotides (~ 4 CG/100 bp). Perhaps consistent with the moderately high CG content of this putative regulatory region, the IG1 transcript was more highly induced in a metl mutant than in drd1 plants (Fig. 2). The fine dotted line and small converging arrows indicate the ~ 400 bp region analyzed by ChIP. Bottom: the short RNA-encoding region is duplicated (90% sequence similarity) on chromosome 3, downstream of a gene encoding the ribosomal protein RPL34C (At3g28900). RT-PCR revealed that a transcript (IG1*; black bar), which overlaps with that detected by whole genome arrays from callus (http://signal.salk.edu/cgi-bin/atta), is also induced in the drd1 mutant. Similarly to IG1, the IG1* region encodes numerous short RNAs of both sense and antisense polarity (Lu et al., 2005).

(B) The intergenic region on chromosome 3 giving rise to the IG2 transcript that is induced in the drd1 mutant. The IG2 transcript detected by cDNA-AFLP (black bar, direction of transcription deduced from 3’ RACE indicated by arrow) is in an intergenic region upstream, and in opposite orientation of, an Athila solo LTR that is truncated at the 3’ end. Short RNAs derived from the solo LTR are indicated by vertical white arrowheads (Lu et al., 2005). The cDNA-AFLP transcript overlaps with a full-length cDNA (light gray bar) from the Riken collection (RAFL08-13-D16; URL in legend to Suppl. Fig. 5) and a transcript detected by whole genome array (dark gray bar) (http://signal.salk.edu/cgi-bin/atta). This transcript is expressed in most organs, which is consistent with 10 ESTs for this sequence in the TAIR database (Suppl. Table I). Because of its opposite orientation relative to the IG2 transcript, the Athila solo LTR presumably acts as an enhancer to boost IG2 expression in the drd1 mutant,
similarly to the LTRCO solo LTR that enhances transcription of the gene encoding the ribosomal protein RPL18C (Fig. 1). The fine dotted line and converging arrows indicate an ~311 bp region encompassing the *Athila* LTR that was analyzed by ChIP. The *Athila* solo LTR is ~220 bp in length and contains 7 CG dinucleotides (~3 CG/100 bp). Perhaps consistent with the moderate CG content, the *IG2* transcript was induced slightly more in the *drd1* mutant than in *met1* plants (Fig. 2).

(C) The intergenic region on chromosome 3 giving rise to the *IG5* transcript that is induced in the *drd1* mutant. The transcript detected by cDNA-AFLP (black bar, direction of transcription indicated by arrow) apparently initiates in the 3’ LTR of an intact *Copia*-like retrotransposon, which is in the same orientation as the *IG5* transcript. Short RNAs derived from the *Copia* LTR are indicated as vertical white arrowheads (Lu et al., 2005). A transcript detected by whole genome array (dark gray bar) is found primarily in callus (http://signal.salk.edu/cgi-bin/atta). The fine dotted line and converging arrows indicate the ~297 bp region in the *Copia* LTR analyzed by ChIP. This sequence contains 17 CG dinucleotides (~6 CG/100 bp). Perhaps consistent with the relatively high CG content, the *IG5* transcript was induced to a greater extent in a *met1* mutant than in *drd1* plants (Fig. 2). Accordingly, CG methylation as assessed by digestion with *Hpa*II and *Msp*I (recognition sequence CCGG; three sites in the ~310 bp region) was reduced in the *met1* mutant (Fig. 3). The three black arrowheads within the LTR denote a tandemly repeated sequence.