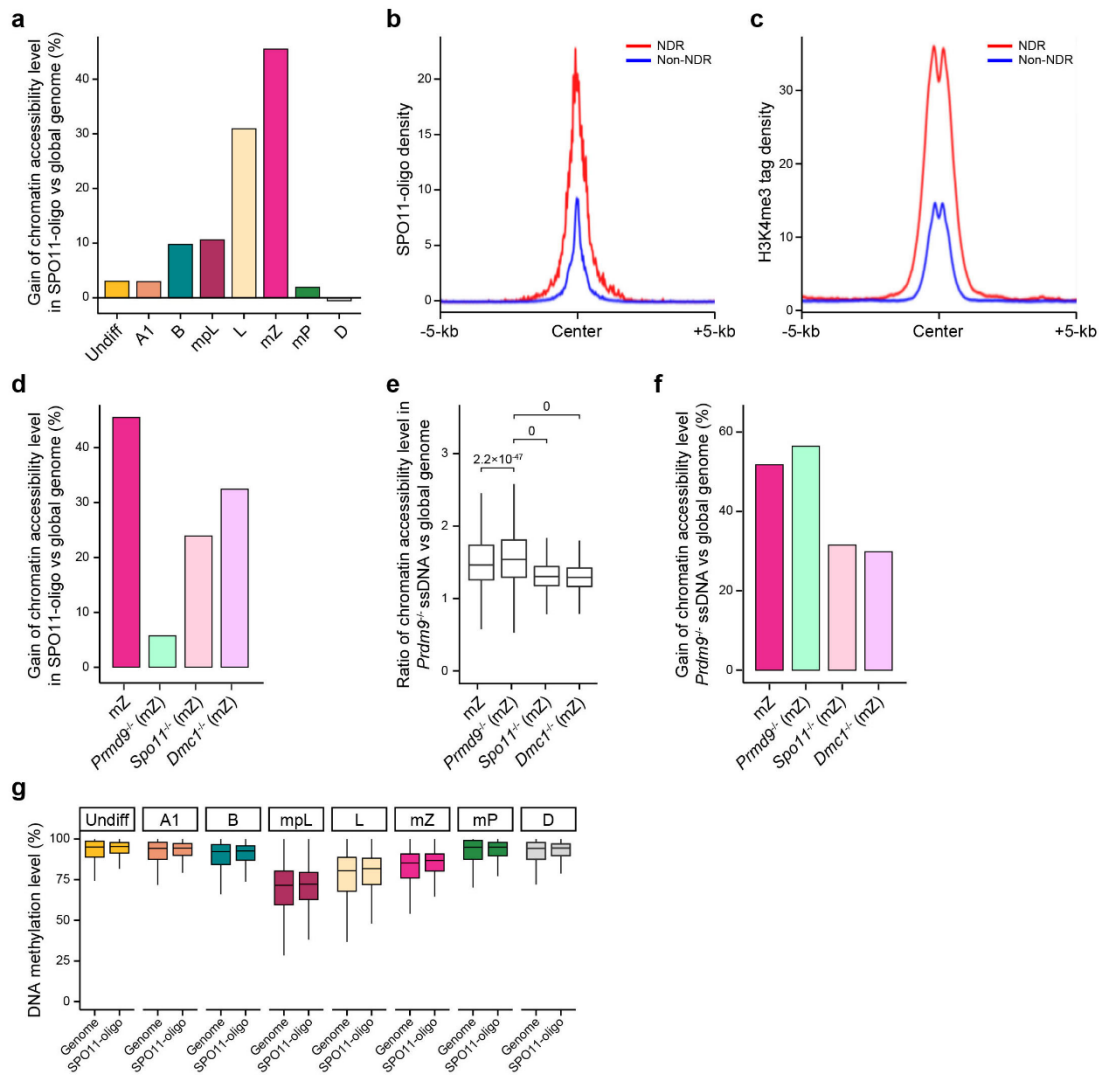


## Supplementary information, Figure S13



**Fig. S13 Chromatin accessibility of the hotspots are positively correlated with DSB**

**hotspots and H3K4me3 signals.** **a** Histogram showing the gain of chromatin

accessibility level (chromatin accessibility level in SPO11-oligo defined DSB hotspots

versus that in the whole genome) in different stages of spermatogenic cells. Only

regions with at least three GCH sites were considered. **b** Profile of the average SPO11-

oligo density on SPO11-oligo defined DSB hotspots with NDRs and without NDRs. **c**

Profile of the average H3K4me3 density on SPO11-oligo defined DSB hotspots with

NDRs and without NDRs. **d** Histogram showing the gain of chromatin accessibility level (chromatin accessibility level in SPO11-oligo defined DSB hotspots versus that in the whole genome) in the wild-type, *Prdm9*<sup>-/-</sup>, *Spo11*<sup>-/-</sup> and *Dmc1*<sup>-/-</sup> mid-zygotene spermatocytes. Only regions with at least three GCH sites were considered. **e** Boxplot showing the ratio of chromatin accessibility level (chromatin accessibility level in *Prdm9*<sup>-/-</sup> DMC1-ssDNA defined DSB hotspots versus that in the whole genome) in the wild-type, *Prdm9*<sup>-/-</sup>, *Spo11*<sup>-/-</sup>, and *Dmc1*<sup>-/-</sup> mid-zygotene spermatocytes. Only regions with at least three GCH sites were considered. **f** Histogram showing the gain of chromatin accessibility level (chromatin accessibility level in *Prdm9*<sup>-/-</sup> DMC1-ssDNA defined DSB hotspots versus that in the whole genome) in the wild-type, *Prdm9*<sup>-/-</sup>, *Spo11*<sup>-/-</sup>, and *Dmc1*<sup>-/-</sup> mid-zygotene spermatocytes. Only regions with at least three GCH sites were considered. **g** DNA methylation level in SPO11-oligo defined DSB hotspots and the whole genome. DSB hotspots were extended as center  $\pm$  2 kb and the whole genome was separated into several tiles with 1-kb size to be calculated the mean DNA methylation. Only regions with at least three WCG sites were considered. Undiff: undifferentiated spermatogonia, A1: type A1 spermatogonia, B: type B spermatogonia, mpL: mid-preleptotene, L: leptotene, mZ: mid-zygotene, mP: mid-pachytene, D: diplotene.