Bumblebees queens health and behaviour are key factors for the production of a new colony. Through their lifetime, bumblebees queens undergo major physiological changes that drive different behaviours and mark important life stages. The molecular basis of these major transitions is largely unknown. In this project, we used whole-genome transcriptomics to characterize the dynamics of gene expression in *Bombus terrestris* queens across key periods of their life: from virgin to mated, overwintered and fully reproductive. Mating occurs with only one male at the end of the summer and triggers important physiological changes that prepare the queen for winter diapause: this lasts several months and is spent underground. Successfully overwintered females emerge at the beginning of the spring, actively forage for pollen and nectar and start a new colony alone. Mating and diapause are highly interconnected (non-mated females are not able to overwinter) and both processes are necessary for the successful production of a new colony in the spring. We sampled bumblebees females from commercial colonies and we induced mating and diapause in standard lab conditions. We obtained four experimental groups of bees: virgin, mated, overwintered with undeveloped ovaries (i.e. non-fertile) and overwintered fully reproductive bees. We dissected brains from 33 bees and we processed these samples for TruSeq mRNA sequencing. We used five lanes of an Illumina Sequencer to produce ~50 million reads per sample with 10X mean coverage. RNA sequencing data were analysed with edgeR for R to identify patterns of differential gene expression across the four experimental groups and we performed Gene Ontology analyses to characterize the biological processes involved in mating behaviour and diapause. These findings provide new insights to understand the molecular basis for fundamental behaviors in bumblebees and might help in developing strategies to improve the health of this important pollinator.