Glucosinolates (GLS) are a major class of secondary metabolites found in the Brassicaceae family, which includes important Brassica crops and the model plant *Arabidopsis thaliana*. The pattern of synthesis and accumulation of these secondary metabolites is greatly influenced by developmental stage and also by the changing environment (i.e. light, temperature or drought). In recent years, efforts have been focussed on the identification of genomic regions that control GLS structural diversity and content in *Arabidopsis*. Information about loci that control GLS under abiotic stresses is still limited, especially in response to low temperatures. Using the Multiparent Advanced Generation Inter-Cross (MAGIC) population we performed a metabolic quantitative trait loci (mQTLs) analysis looking for GLS variation under low temperatures. The MAGIC population consist in 527 recombinant inbred lines (RILs) descended from a heterogeneous stock of 19 intermated accessions of the plant *Arabidopsis thaliana* which increase the allelic and phenotypic diversity over traditional inbred lines. Experiments were carried out at control (HT, 20 °C) and cold (LT, 10 °C) conditions. We identified 50 mQTLs that regulate the biosynthesis of GLS at HT and 23 at LT. Most of the 23 mQTLs identified at LT, are involved in the synthesis of aliphatic GLS, with the exception of one mQTL regulating the biosynthesis of neoglucobrassicin and two of gluconasturtiin. These 23 mQTLs are placed in 8 independent genomic regions. Two of them, namely C1-12 and C5-23, were identified exclusively at LT. The C5-23 region encompasses three genes associated to GLS synthesis (APO2, ATR1 and MYB28), whereas in the C1-12 region no genes directly involved in GLS metabolism were found, which make this region an interesting target for future research.