Background Growing evidence are demonstrating the therapeutic efficacy of immune checkpoint inhibitors (ICI) in mesothelioma; however, a limited percentage of patients benefits from this therapeutic approach. Epigenetic modifications play a relevant role in negatively regulating the cross-talk between neo-plastic and immune cells, and in contributing to the highly immunosuppressive mesothelioma microenvironment. A better understanding of mesothelioma epigenetic landscape could open the path to novel and potentially more effective approaches combining ICI and epigenetic drugs. We investigated the immunomodulatory potential of epigenetic agents by comparing the activity of DNA hypomethylating agents (DHA) with histone deacetylases inhibitors (HDACi) and EZH2 inhibitors (EZH2i), alone or combined with DHA, in mesothelioma cells. Methods Four mesothelioma cell lines were treated with the DHA guadecitabine 1µM, or with the HDACi, Valproic Acid (VPA) 1mM, or the EZH2i, EPZ-6438 1µM, alone or combined with guadecitabine. We investigated the expression of HLA class I molecules by flow-cytometry and of PD-L1, cancer testis antigens (CTA: NY-ESO, MAGE-A1), Natural Killer Group 2 member D Ligands (NK2DLS: MIC-A, MIC-B, ULBP2) and EMT-regulating cadherins (CDH1, CDH2) by quantitative Real-Time PCR. Fold change (FC) expression for each treatment vs untreated cells was reported as mean values (FCm) among investigated cell lines. A positive modulation of the expression was considered if FCm>1.5. Results Guadecitabine upregulated the expression of HLA class I antigens (FCm=1.75), PD-L1 (FCm=2.38), NK2DLS (MIC-A FCm=1.96, MIC-B FCm=2.57, and ULBP2 FCm=3.56), and upregulated/induced CTA expression. Similarly, VPA upregulated HLA class I antigens (FCm=1.67), PD-L1 (FCm=3.17), NK2DLS (MIC-A FCm=1.78, MIC-B FCm=3.04, and ULBP2 FCm=3.75) expression; however, CTA expression was modulated only in 1 mesothelioma cell line. Conversely, EPZ-6438 up-regulated only NY-ESO-1 and MIC-B expression in 1 mesothelioma cell line. The addition of both VPA and EPZ-6438 to guadecitabine strengthened its immunomodulatory activity. Specifically, guadecitabine plus VPA or EPZ-6438 upregulated the expression of HLA class I antigens FCm=2.53 or 2.69, PD-L1 FCm=8.04 or 2.65, MIC-A FCm=3.81 or 2.26, MIC-B FCm=8.00 or 3.03, ULBP2 FCm=6.24 or 4.53, respectively. Higher levels of CTA upregulation/induction were observed with combination treatments vs guadecitabine alone. Cadherins modulation was mesothelioma histotype-related: CDH1 expression was induced in the 2 constitutively-negative sarcomatoid mesothelioma cells by guadecitabine alone or combined with VPA or EPZ-6438; CDH2 expression was upregulated by VPA alone (FCm=1.53) or plus guadecitabine (FCm=2.54). Conclusions Combination of DHA-based immunotherapies with other classes of epigenetic drugs could be an effective strategy to be pursued in the mesothelioma clinic.