

Supporting information

Table S1. Oligonucleotide sequences used for qPCR

Gene	Forward	Reverse
UL123	GCCTTCCCTAAGACCACCAA	ATTTTCTGGGCATAAGCCATAATC
UL55	TCAAGGTGCTGCGTGATATG	CGAGCTGTTGGCGAAATTAAAG
GAPDH	GGTGTGAACCATGAGAAGTATGA	GAGTCCTTCCACGATACCAAAG

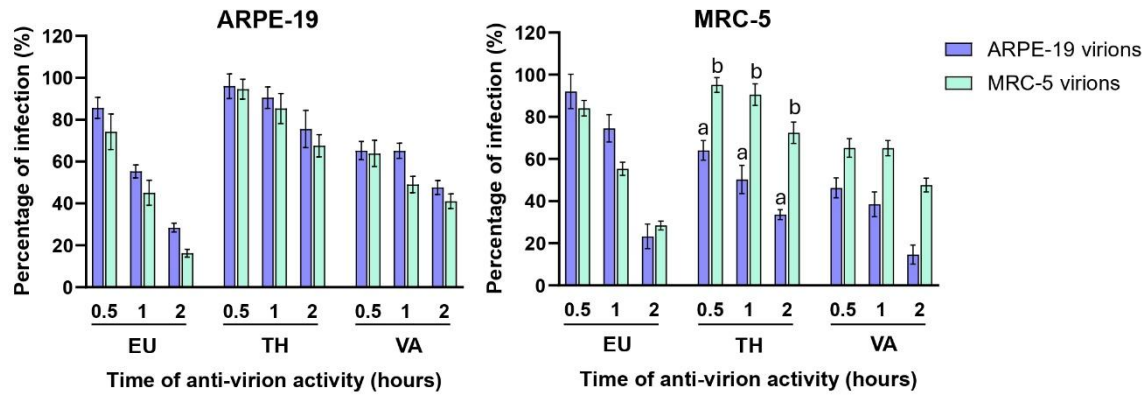


Fig. S1. Anti-virion activity of EOCs against HCMV virions produced from ARPE-19 and MRC-5 cells, measured as the percentage of infection obtained in ARPE-19 or MRC-5 cells. Both ARPE-19-derived and MRC-5-derived virions were incubated at 37°C for 0.5, 1 or 2 hours with DMEM containing the EC₅₀ concentration (obtained in the antiviral assay) of EOCs prior to inoculation of these virions. The percentage of infection was calculated relative to DMSO treatment. Statistical significance between ARPE-19 virions and MRC-5 virions was determined by a nested t test. Different letters above bars indicate significant differences between groups ($p < 0.05$). Bars labeled with different letters (a, b) present significant differences.

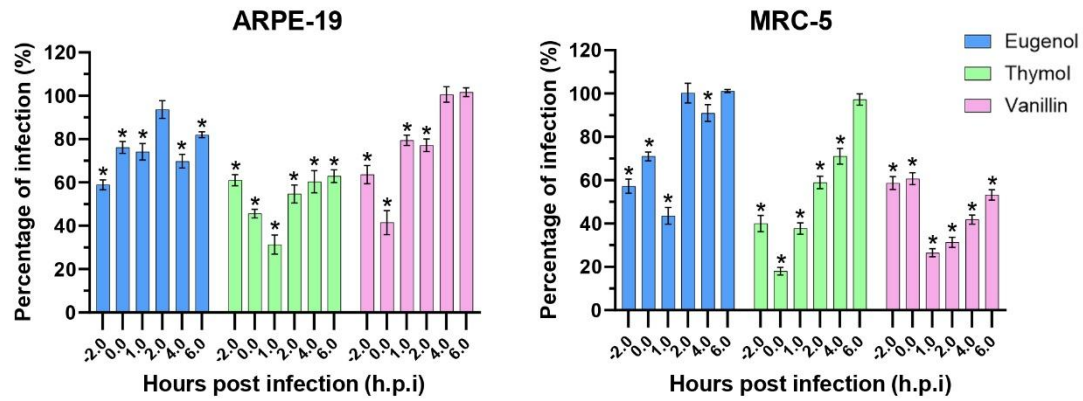


Fig. S2. Time-of-addition assay. ARPE-19 and MRC5 cells were infected and treated with the EC₅₀ of the EOCs at various times relative to infection (-2, 0, 1, 2, 4, 6 hpi). Data represents mean values (\pm SD). Percentage of infection was determined by using DMSO-treated cells as 100%. The error bars represent the standard deviation from six replicates. Significance was determined by performing a two-tailed t-test with comparisons to infected DMSO-treated cells as a control. Data represents the mean \pm standard deviation of six biological replicates.

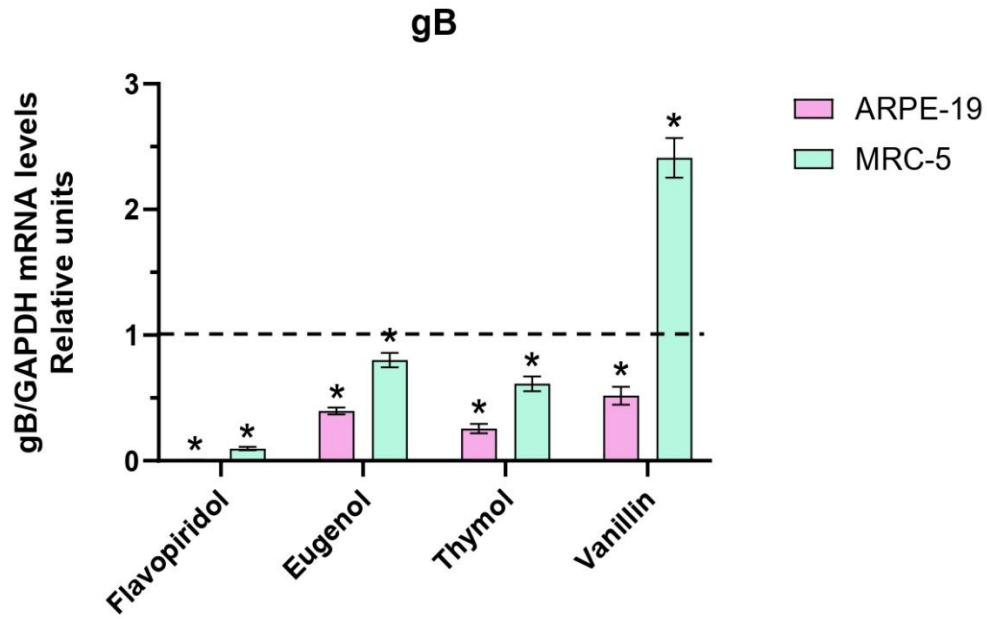


Fig. S3. Effects of eugenol, thymol and vanillin on HCMV gene expression. Depicted are mRNA levels of the gB and GAPDH genes as quantified by qPCR. Data represents mean values (\pm SD). Dashed line indicates the expression of the control condition. The asterisk (*) denotes significant differences compared to DMSO-treated cells.

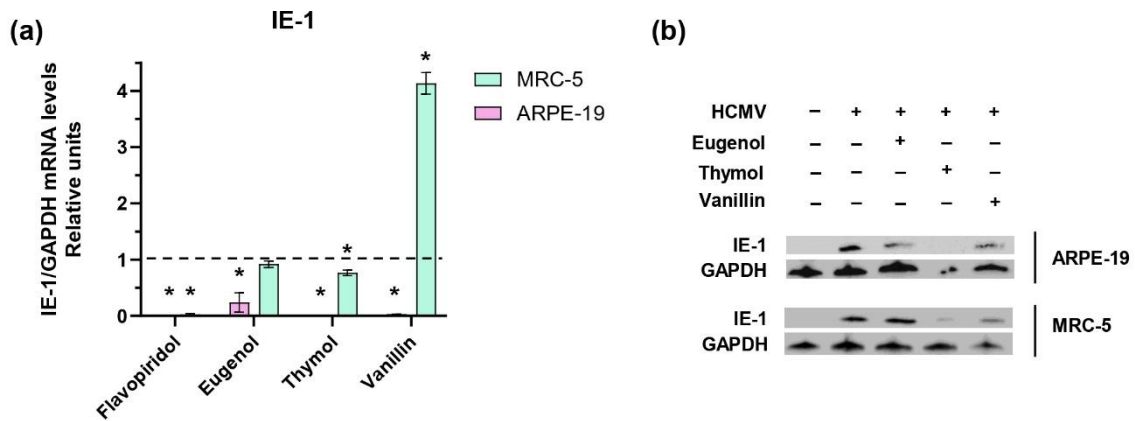


Fig. S4. ARPE-19 and MRC-5 cells were treated for 2 h with the EC₉₀ of EOCs, followed by infection. Subsequently, the cultures were incubated for 48 hours at 37°C, after which the cells were harvested by scraping. (a) IE-1 and GAPDH mRNA levels quantified by qPCR are presented. Flavopiridol was used as a positive control for mRNA synthesis inhibition. Dashed line indicates the expression of the control condition. The asterisk (*) denotes significant differences compared to DMSO-treated cells. (b) HCMV protein levels in EOCs-treated cells. Cells were lysed after 48 hours and protein lysate analyzed by Western blot using the indicated antibodies. GAPDH level was used as a loading control. In ARPE-19 cells, TH at its EC₉₀ concentration is highly toxic, reducing cell viability to approximately 20% (Fig. 1c). Consequently, we had to load 3 µg of protein, which likely affected the appearance of the loading control.

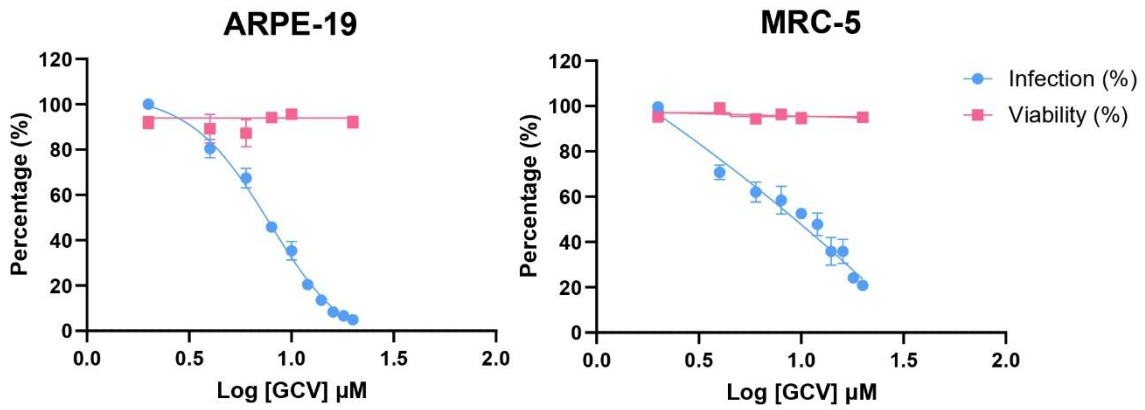


Fig. S5. Percentage of viability and infection after treatment with GCV in ARPE-19 and MRC-5 cells according to the logarithm of the EOC concentration. HCMV infected cells (after 12h for MRC-5 and 24h for ARPE-19) were treated with various concentrations (0-20 μM) of GCV. At 3- and 6-dpi, media and media compound mixture were replaced. At 7 dpi, cells were fixed in 1X PFA and the plates were analyzed with a fluorescence microscope (Cytell) to determine the number of GFP-positive cells relative to the positive control (DMSO 0.1%).

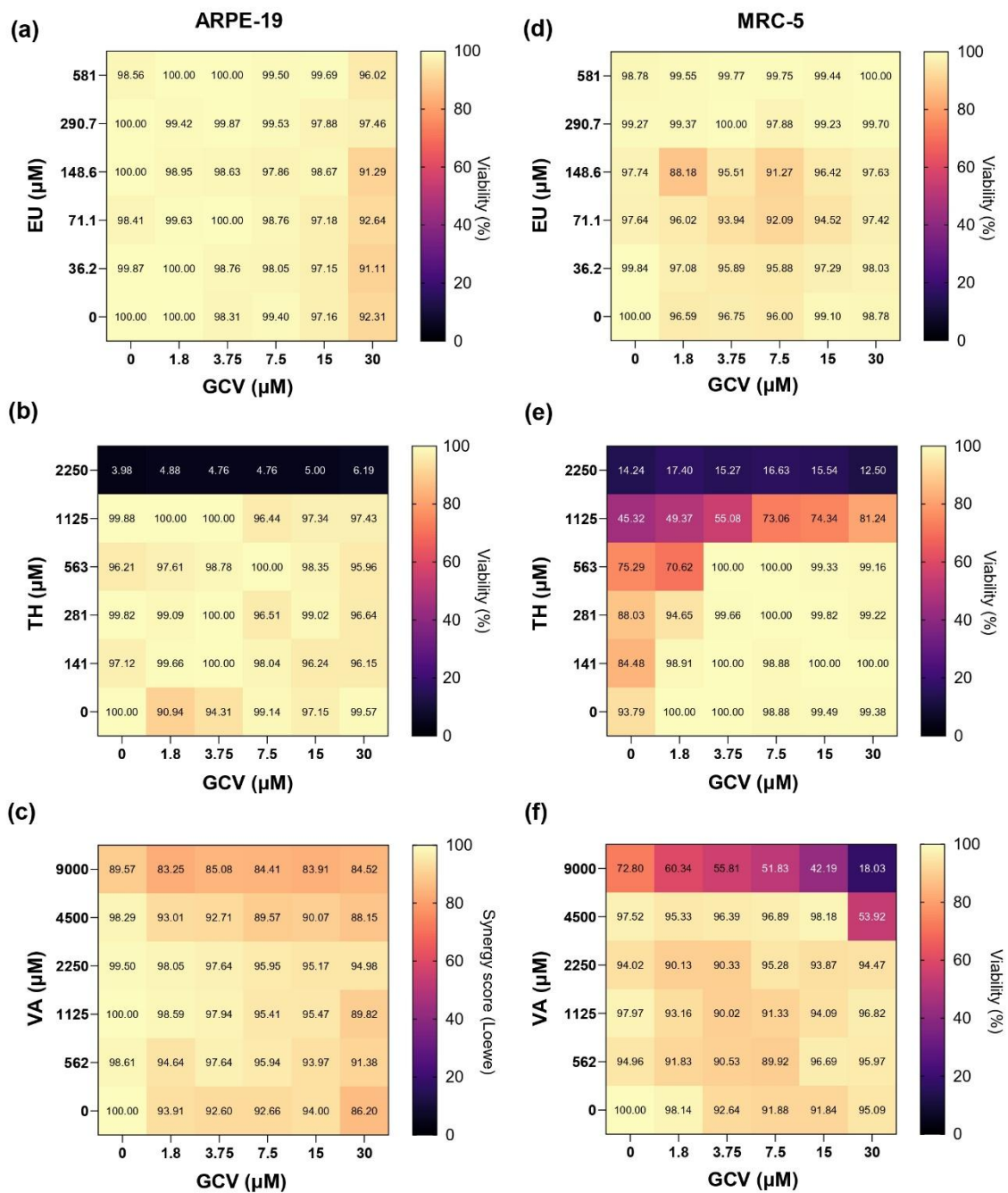


Fig. S6. Checkerboard assay evaluating the percentage of cell viability in ARPE-19 cells (a, b, c) and MRC-5 cells (d, e, f) treated with EOCs-GCV, assessed using the AlamarBlue assay. Color intensity reflects cell viability, with lighter shades indicating higher viability and darker shades indicating lower viability.