Supporting information

Table S1: Properties of small molecules and antibodies used in the present study.

Compound/ mAb	Activity in cell-based adhesion assays (IC_{50} shown if not otherwise indicated)	Reference	Comments
XVA143	0.7 nM	(Mancuso, Welzenbach, Steinberger, Krahenbuhl & Weitz-Schmidt, 2016)	Concentration used 10 μM
LFA878	96 nM	(Mancuso, Welzenbach, Steinberger, Krahenbuhl & Weitz-Schmidt, 2016)	Concentration used 10 μM
BIRT377	26 nM (Kd value)	(Kelly et al., 1999)	Concentration used 10 μM
Firategrast	na	na	Based on IC50 values published for derivatives, the IC50 of firategrast is assumed to be in the low nM range (Baiula); concentration used 10 μM
RO0505376	11 nM	Mancuso, unpublished data	Concentration used 10 μM
Lifitegrast	3 nM	Mancuso, unpublished data	Concentration used 10 μM
mAb R7.1	10 μg mL^{-1} results in 70% inhibition	(Lorenz, Harrer, Lagoo, Baur, Eger & Kalden, 1993)	R7.1 is commonly used at 10 μg mL^{-1} in functional studies; concentration used 10 μg mL^{-1}
mAb TS1/22	5 μg mL^{-1} results in complete inhibition	(Petruzzelli, Maduzia & Springer, 1998)	Concentration used 10 μg mL^{-1}
Efalizumab	0.06 μg mL^{-1}	(Mancuso, Welzenbach, Steinberger, Krahenbuhl & Weitz-Schmidt, 2016)	Therapeutic serum levels: 10 μg mL^{-1} (Guttman-Yassky et al., 2008); concentration used 10 μg mL^{-1}
Natalizumab	10 μg mL^{-1} =saturating amount	(Sehr et al., 2016)	Trough serum levels: MS patients: 29 μg mL^{-1} Crohn's patients: 10 μg mL^{-1} (Ref.: described in US package insert Tysabri); concentration used 10 μg mL^{-1}
Vedolizumab	0.02–0.06 μg mL^{-1}	(Soler, Chapman, Yang, Wyant, Egan & Fedyk, 2009)	Trough serum levels: Ulcerative colitis: 26.3 μg mL^{-1} (week 0-6); 11 μg mL^{-1} (week >6) Crohn's disease: 27.4 μg mL^{-1} (week 0-6); 13 μg mL^{-1} (week >6) (Ref. US package insert Entyvio); concentration used 10 μg mL^{-1}

mAb: monoclonal antibody; na: not available

References cited in Table S1

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Soler D, Chapman T, Yang LL, Wyant T, Egan R, & Fedyk ER (2009). The binding specificity and selective antagonism of vedolizumab, an anti-alpha4beta7 integrin therapeutic antibody in development for inflammatory bowel diseases. J Pharmacol Exp Ther 330**:** 864-875.

Figure S1

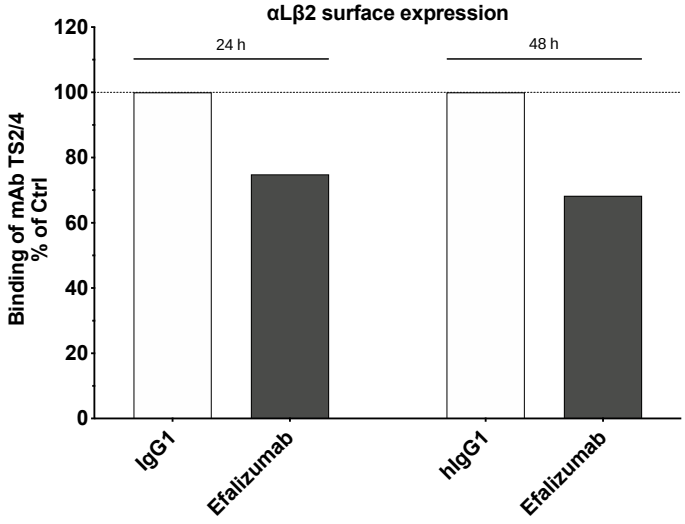

Figure S1: Effect of efalizumab on integrin αLβ2 surface expression. Surface expression of αLβ2 on CD2+ T cells after 24 hours and 48 hours of treatment with anti-αLβ2 efalizumab (10 μg mL^{-1}), and respective isotype control (hIgG1). Each bar represents a single determination using blood from one donor.

Figure S2:

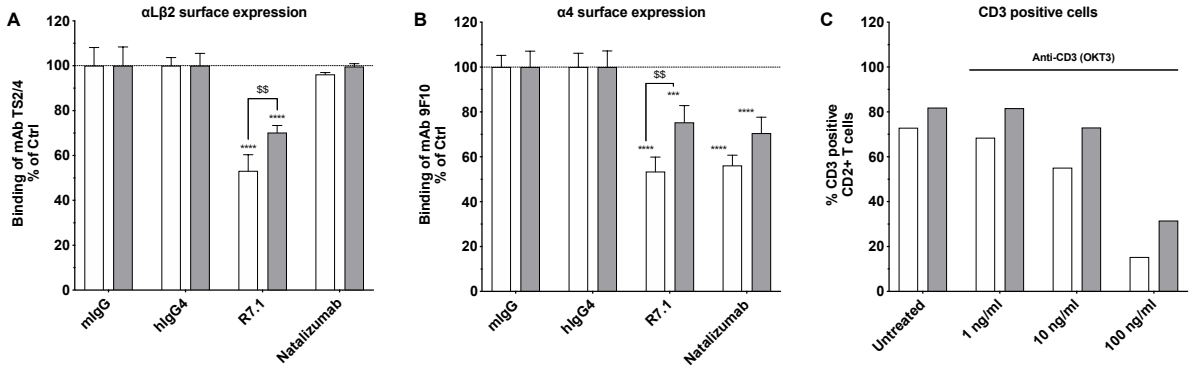

FIGURE S2: Effect of dynasore on mAb R7.1-induced αLβ2 and α4 downmodulation, natalizumab-induced α4 downmodulation and mAb OKT3-induced downmodulation of CD3. Surface expression of **(A)** αLβ2 and **(B)** α4 integrin on CD2+ T cells after 24 hours of treatment with anti-αL R7.1 (10 μg mL^{-1}), anti-α4 natalizumab (10 μg mL^{-1}), and respective isotype controls (mIgG1, hIgG4). Treatments were performed in the presence of dynasore (40 μM, grey bars) or DMSO (white bars). Each bar represents the mean value ± SEM of 6 independent experiments using blood samples from different donors. Statistical significance was determined by using one-way ANOVA, ***$P < 0.001$, ****$P < 0.0001$ *vs* control, and two-way ANOVA, $^{\$\$}P < 0.01$ *vs* incubation without dynasore. **(C)** Surface expression of CD3 on CD2+ T cells after 24 hours of treatment with anti-CD3 OKT3 (1 ng mL^{-1}, 10 ng mL^{-1}, 100 ng mL^{-1}) in the presence of dynasore (40 μM, grey bars) or DMSO (white bars). Each bar represents a single determination, using blood from one donor.