Supplementary Materials 1

Radioligand binding assay protocol
For detection of α_{1A}-adrenergic receptor (AR) activation, submaxillary gland samplesof male Wistar rats were homogenized in modified Tris-HCl buffer (pH 7.4) using standard techniques, and aliquots of the prepared samples was incubated with 0.25 nM [^{3}H] prazosin for 60 min at 25°C. Nonspecific binding was estimated in the presence of 10 µM phentolamine (Michel et al., 1989). For detection of α_{1B}-AR activation, liver samplesof male Wistar rats were prepared in modified Tris-HCl buffer using standard techniques and aliquots of the sample/Tris-HCl homogenate was incubated with 0.25 nM [^{3}H] prazosin for 60 min at 25°C. Nonspecific binding was estimated in the presence of 10 µM phentolamine (Garcia-Sainz et al., 1992; Michel et al., 1989). For the α_{2A}-AR binding assay, insect Sf9 cells expressing human recombinant α_{2A}-AR were homogenized in modified Tris-HCl buffer and aliquots were incubated with 1 nM [^{3}H] MK-912 for 60 min at 25°C. Nonspecific binding was estimated in the presence of 10 µM WB-4101 (Uhlen et al., 1994). For α_{2B}-AR, CHO-K1 cells stably transfected with a plasmid encoding human α_{2B}-AR were homogenized in modified Tris-HCl buffer using standard techniques and aliquots were incubated with 2.5 nM [^{3}H] rauwolscine for 60 min at 25°C. Nonspecific binding was estimated in the presence of 10 µM prazosin (Uhlen et al., 1998). For α_{2C} -AR, insect Sf9 cells expressing human recombinant α_{2C}-AR were homogenized in modified Tris-HCl buffer and aliquot was incubated with 1 nM [^{3}H] MK-912 for 60 min at 25°C. Nonspecific binding was estimated in the presence of 10 µM WB-4101 (Uhlen et al., 1994). For β_1 -AR, CHO-K1 cells expressing human recombinant β_1-AR were homogenized in modified Tris-HCl buffer and aliquots were incubated with 0.03 nM [^{125}I] cyanopindolol for 120 min at 25ºC. Nonspecific binding was estimated in the presence of 100 µM S (-)-propranolol (Feve et al., 1994). For β_3-AR, HEK-293 cells expressing human β_3-AR expressed were used to prepare membranes in modified Tris-HCl buffer and aliquots were incubated with 0.5 nM [^{125}I] iodocyanopindolol for 90 min at 25°C. Nonspecific binding was estimated in the presence of 1 mM alprenolol (Feve et al., 1994). All samples were filtered twice and assayed to determine radio ligand-specific binding. The half maximal inhibitory concentrations (IC_{50}) values were determined by nonlinear, least-squares regression analysis using MathIQ™ statistical software (ID Business Solutions Ltd., Surrey, UK).

References

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