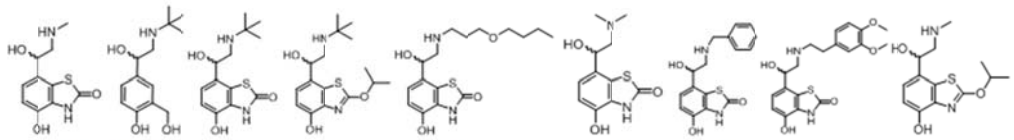
The effect of alterations to the *N*-alkyl moiety and benzothiazolone on affinity, efficacy and selectivity of the β_2-agonist S1319 at the human β_1- and β_2-adrenoceptors

S1319 was originally isolated from the marine sponge *Dysidea sp.* and has several structural similarities to adrenaline and salbutamol. It has been reported to have affinities of 120nM and 50nM for the β_1 and β_2-adrenoceptor(AR)s respectively but very high potency (log EC_{50} -10.6) for β_2-mediated tracheal relaxation compared to salbutamol (Suzuki, et al., 1999a and b). The aim of this study was to investigate whether alterations to the *N*-alkyl group and the addition of an isopropyl group on the benzothiazolone affected the affinity, efficacy or selectivity of S1319 derivatives for the human β_1 and β_2-ARs.

In this study, the parent compound S1319 and several analogues were synthesized. These were investigated using ^{3}H-CGP12177 whole cell binding and CRE-SPAP gene transcription in CHO cells stably expressing a CRE-SPAP reporter gene and either the human β_1 or β_2-AR (Baker, 2010a and b).



S1319 salbutamol **1** **2** **3** **4** **5** **6** **7**

	Human β_1-adrenoceptor					Human β_2-adrenoceptor				
	Log K_D	n	Log EC_{50}	%isop max	n	Log K_D	n	Log EC_{50}	% isop max	n
S1319	-5.50±0.06	6	-6.76±0.14	93.5±6.8	9	-7.37±0.06	7	-9.33±0.14	97.6±3.4	10
salbutamol	-4.61±0.05	5	-6.57±0.10	78.3±2.9	5	-5.97±0.05	4	-8.43±0.08	92.3±4.7	6
1	-7.09±0.06	7	-9.06±0.11	95.5±3.6	12	-9.25±0.09	7	-10.02±0.13	106.6±3.7	12
2	-6.05±0.03	8	-7.88±0.13	89.3±2.3	12	-8.00±0.04	8	-8.98±0.10	98.6±3.6	11
3	-6.70±0.06	8	-8.16±0.14	99.0±3.8	13	-7.58±0.05	7	-9.67±0.11	101.2±4.8	10
4	-4.73±0.11	5	-6.29±0.07	95.7±6.6	10	-6.29±0.08	9	-8.50±0.16	98.8±5.2	10
5	-5.73±0.12	6	-7.35±0.18	96.7±8.1	8	-7.49±0.10	6	-9.93±0.11	103.8±4.2	7
6	-7.50±0.05	5	-8.79±0.20	89.4±2.0	6	-7.49±0.09	6	-9.50±0.22	97.6±4.4	7
7	-4.77±0.06	5	-6.09±0.16	90.6±5.9	11	-6.33±0.07	8	-8.72±0.14	94.1±4.0	11

Table. Log K_D values from ^{3}H-CGP12177 binding and log EC_{50} values and % maximum isoprenaline responses from CRE-SPAP production. Values are mean±sem of n separate experiments.

S1319 was confirmed to be a β_2-selective ligand with a 74-fold increase in affinity for the β_2-AR over the β_1-AR. It was also confirmed to be a full agonist at both receptors, with high potency for the β_2-AR. The addition of the *tert*-butyl group (**1**) increased the affinity of the molecule for both receptors by 30- to 100- fold, however the addition of an ether side-chain (**3**) or benzyl substituent (**5**) had little effect on the affinity or efficacy. Alteration of the benzothiazolone head group to the isopropoxy analogue (**2, 7**) decreased the affinity and potency at both receptors relative to their parent compounds (**1**, S1319). All of the synthesised ligands retained full agonism compared with isoprenaline.

Baker JG (2010a) *Br. J. Pharmacol* **160:** 148–161.
Baker JG (2010b) *PLoS ONE* 5(11): e15487.
Suzuki et al., (1999a) *Bioorganic and Med Chem Lett* 9: 1361–1364.
Suzuki et al., (1999b) *Br. J. Pharmacol* **128:** 716–720.