G-protein-coupled receptors (GPCRs) are essential components of the signalling network throughout the body. To understand the molecular mechanism of G-protein-mediated signalling, solved structures of receptors in inactive conformations and in the active conformation coupled to a G protein are necessary. Here we present the structure of the adenosine A2A receptor (A2AR) bound to an engineered G protein, mini-Gα, at 3.4 Å resolution. Mini-Gα binds to A2AR through an extensive interface (1,048 Å²) that is similar, but not identical, to the interface between Gs and the β2-adrenergic receptor. The transition of the receptor from an agonist-bound active-intermediate state to an active G-protein-bound state is characterized by a 14 Å shift of the cytoplasmic end of transmembrane helix 6 (H6) away from the receptor core, slight changes in the positions of the cytoplasmic ends of H5 and H7 and rotamer changes of the amino acid side chains Arg3.50, Tyr5.58 and Tyr7.53. There are no substantial differences in the extracellular half of the receptor around the ligand binding pocket. The A2AR−mini-Gα structure highlights both the diversity and similarity in G-protein coupling to GPCRs and hints at the potential complexity of the molecular basis for G-protein specificity.

Structures of A2AR bound to either inverse agonists or agonists have elucidated the molecular determinants of subtype specificity and ligand efficacy. However, the mechanism of activation of the receptor to allow G-protein coupling and the basis of G-protein selectivity is not fully understood. Structures of A2AR in the inactive state have been determined bound to the antagonists ZM241385 (refs 7–9), XAC7, caffeine or 1,2,4-triazines, and all the structures are very similar. An intramembrane Na⁺ ion that can act as an allosteric antagonist was identified in the highest resolution structure (1.8 Å) (ref. 13), and a homologous Na⁺ ion has been subsequently identified in other high-resolution structures of GPCRs. Four agonist-bound structures of A2AR have also been determined after co-crystallization with either adenosine, NECA, CGS21680 (ref. 10) or UK432097 (ref. 5). All the structures are very similar and are thought to represent an active-intermediate conformation of the receptor, but not the fully active receptor that binds a G protein. Observations that support this conclusion include the presence of rotamer changes of conserved amino acid residues associated with activation of other GPCRs, and the absence of a large-scale movement of the cytoplasmic end of transmembrane helix 6 (H6) away from the receptor core. The G-protein-coupled state of A2AR exhibits higher binding affinity for agonists compared to the uncoupled state, but it is unclear whether the agonist-bound structures determined so far depict the binding pocket in a high-affinity or low-affinity conformation. Therefore, in order to elucidate the structure of the activated state of A2AR, we have determined its structure bound to a high-affinity agonist and an engineered G protein.

There is a single reported structure of a GPCR bound to a heterotrimeric G protein, namely Gαi-bound β2-adrenergic receptor (β2AR)3, which showed that virtually all the atomic contacts between the receptor and G protein were formed by the Goi subunit. To facilitate the crystallization of any GPCR–Gs complex, we developed a minimal G protein, mini-Gα, that comprised a truncated form of the GTase domain of Goα, and included eight point mutations to stabilize the protein in the absence of Gβγ and in the presence of detergents (B.C. and C.G.T., manuscript submitted). In addition, three truncations removed the switch III region, 25 amino acids from the N terminus and the α-helical domain. Mini-Gα reproduced the

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**Figure 1 | Ligand binding and overall structure of the A2AR−mini-Gα complex.**

[a] The structure of A2AR is depicted as a cartoon in rainbow coloration (N terminus in blue, C terminus in red) with mini-Gα in purple. The agonist NECA bound to A2AR and GDP bound to mini-Gα are depicted as space-filling models (carbon, yellow; nitrogen, blue; oxygen, red; phosphorous, orange). Relevant secondary structural features are labelled. b. Mini-Gα increases the affinity of agonist binding to A2AR similar to that observed by a heterotrimeric G protein. Competition binding curves were performed in duplicate (n = 3) by measuring the displacement of the inverse agonist 3H-ZM241385 with increasing concentrations of the agonist NECA (K values in parentheses, see Extended Data Fig. 1 for full data): blue circles, A2AR (K = 46 ± 0.34 µM); orange squares, A2AR and mini-Gα (K = 430 ± 80 nM); green diamonds, A2AR and heterotrimeric G protein with nanobody Nb35 (K = 340 ± 70 nM). Error bars represent s.e.m. G proteins were all added to membranes containing A2AR to give a final concentration of 25 µM and the final concentration of NaCl was 100 mM (see Methods). c. The structure of β2AR (green) bound to Goi (grey and purple) is depicted as a cartoon in the same orientation as A2AR in a; the purple region in Goi corresponds to the structure of mini-Gα.
increase in agonist affinity that occurred upon incubation of the receptor in the presence of the heterotrimeric G protein Gs and it also showed identical sensitivity to the presence of the allosteric antagonist Na\(^+\) (Fig. 1 and Extended Data Fig. 1). In addition, mini-Gs readily formed a complex with A\(_{2a}\)R in the presence of the agonist NECA and the complex was considerably more thermostable, particularly in...
short-chain detergents, than A2AR with only NECA bound (Extended Data Fig. 2). This complex was crystallized in the detergent octylthioglucoside by vapour diffusion, a data set collected from two crystals, the structure determined by molecular replacement (see Methods) and refined to 3.4 Å (Extended Data Table 1). Of the two A2AR–mini-Gs complexes per crystallographic asymmetric unit, the determination with Arg2917.56 and forms the sequence different amino acid sequences at the H7–H8 boundary. In A2AR, H7 (Extended Data Fig. 7). The main difference between the A2AR–mini-Gs atomic contacts may differ (Fig. 3, Extended Data Fig. 6, Supplementary and the receptors are conserved, although the exact orientation and in A2AR and another ten present only in β2AR (Fig. 3 and Extended Data Fig. 5), which results in a slightly different packing of the G proteins to the receptors, although we cannot discount the possible influence of lattice contacts. Alignment of mini-Gs, with Gox, bound to β2AR shows that they are essentially identical (r.m.s.d. 0.92 Å over 1,158 atoms), with the most substantial difference being an 8° tilt between the respective α5 helices, resulting in a 3.7 Å displacement of the Cα of Tyr391 in mini-Gs away from the core of the G protein (Extended Data Fig. 8). Overall, there are 14 contacting residues in common between the β2AR–Gs complex and the A2AR–mini-Gs complex, with an additional 6 contacting residues present only in A2AR and another ten present only in β2AR (Supplementary Table 1). Many of the contacts between residues in the α5 helix of the G protein and the receptors are conserved, although the exact orientation and atomic contacts may differ (Fig. 3, Extended Data Fig. 6, Supplementary Table 1). Similarly, there is a highly conserved interaction between a hydrophobic residue in the centre of CL2, Leu110 in A2AR and Phe139 in β2AR, and residues His41S1.2, Val217S3.1 and Asp215s2s3.1 in Gox (Extended Data Fig. 7). The main difference between the A2AR–mini-Gs interface compared to the β2AR–Gs interface occurs as a result of the different amino acid sequences at the H7–H8 boundary. In A2AR, H7 terminates with Arg2917.56 and forms the sequence R756REFR (bold

**Figure 4**  Conformational changes in A2AR upon G-protein binding. A2AR (rainbow colouration) bound to mini-Gs (purple) was aligned with A2AR in the active-intermediate conformation bound to either NECA (PDB ID: 2YDV) or UK432097 (PDB ID: 3QAK) to highlight structural changes upon G-protein binding. Neither structure was used for both comparisons because the large extensions of the ligand UK432097 compared to NECA distorts the extracellular surface in comparison to the NECA-bound structure and the NECA-bound structure contains a thermostabilizing mutation in the intracellular half of the receptor. 

a, Alignment between 2YDV and the extracellular half of the A2AR–mini-Gs complex is viewed parallel to the membrane plane.

b, Alignment with 3QAK and viewed from the cytoplasmic surface with mini-Gs removed for clarity. c, Alignment with 3QAK viewed parallel to the membrane with the cytoplasmic side at the bottom. Residues are labelled with their Ballesteros–Weinstein numbers and arrows depict the direction of movement upon mini-Gi binding. Conversion of Ballesteros–Weinstein and CNG numbers to amino acid residues in A2AR and mini-Gs, respectively, are as follows: R756, Arg102; Y558, Tyr197; K629, Lys227; L637, Leu235; Y753, Tyr288; Y1023, Tyr391; L1051, Lys227; A633, Ala231 carbonyl; L637, Leu235; Y753, Tyr288; Y1023, Tyr391; L1051, Lys227;
amino acid residues make contact with mini-G<sub>i</sub>, compared to the sequence S<sup>56</sup>PDFRIR in the equivalent position of β<sub>3</sub>AR, where none of the residues make contacts with G<sub>α</sub><sub>i</sub>. Another region of the receptors that differs in the presence/absence of contacts to their respective G proteins is at the end of H5, owing to the extension of H5 in β<sub>3</sub>AR by an additional turn compared to A<sub>2A</sub>R (Fig. 3, Extended Data Fig. 6 and Supplementary Table 1). From these examples it is clear that although the majority of amino acid residues at the interface between the receptor and G protein are identical, the specific atoms involved in the contacts differ either in terms of the amino acid side chains involved, their relative dispositions at the interface and/or the nature of the interaction.

Comparison of the active-intermediate state of UK432097-bound A<sub>2A</sub>R<sup>5</sup> with the structure of A<sub>2A</sub>R bound to mini-G<sub>i</sub> identified major rearrangements in the cytoplasmic half of the receptor core to accommodate G-protein binding (Fig. 4) and will be described in terms of the rearrangements required to transition from the active-intermediate state to the activated G-protein-bound conformation. First, the cytoplasmic end of H6 moves away from the receptor core by 14 Å as measured between the Cα atoms of Thr224<sup>6.26</sup> and Tyr288<sup>7.53</sup>. This movement is achieved through H6 bending outwards with little discernible rotation around the helix axis. The extent of H6 movement is dictated by van der Waals interactions between Lys227<sup>6.29</sup>, Ala231<sup>6.33</sup> and Leu235<sup>6.37</sup> in A<sub>2A</sub>R and Leu393<sup>14.25</sup> and the carboxy terminus of mini-G<sub>i</sub>. The movement of H6 involves substantial changes in the packing of the cytoplasmic end of H6 with helices H5 and H7. In particular, the side chains of highly conserved Tyr197<sup>5.58</sup> and Tyr288<sup>7.53</sup> both adopt new rotamers to fill the space previously occupied by the side chains of Leu235<sup>6.27</sup> (the Cα of which moves by 3.7 Å) and Ile238<sup>6.46</sup> (Cα moves by 2.2 Å), respectively. The shift in Tyr288<sup>7.53</sup> allows Arg102<sup>3.50</sup> of the conserved DRY motif to adopt a fully extended conformation, packing against the end of H6 with helices H5 and H7. In particular, the side chains of the 2.6 angstrom crystal structure of a human A<sub>2A</sub> adenosine receptor bound to an antagonist. Science 322, 1211–1217 (2009).

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Author Contributions B.C. performed receptor expression, purification, crystallization, crystallography, cryo-cooling of the crystals, data collection, data processing and structure determination. T.W. helped with crystallization, data collection and data processing. R.N. performed the stability assays and pharmacological analyses on A<sub>2A</sub>R-mini-G<sub>i</sub> complexes. A.G.W.L. was involved in data processing and structure solution, refinement and analysis. Manuscript preparation was performed by B.C., A.G.W.L. and C.G.T. The overall project management was by C.G.T.

Author Information Atomic co-ordinates and structure factors for the A<sub>2A</sub>R-mini-G<sub>i</sub> complex have been submitted to the Protein Data Bank (PDB) under accession code 5G53. Reproductions and permissions information is available at www.nature.com/reprints. The authors declare competing financial interests: details are available in the online version of the paper. Readers are welcome to comment on the online version of the paper. Correspondence and requests for materials should be addressed to B.C. (byronc@mrcl-lmb.cam.ac.uk) or C.G.T. (cgt@mrc-lmb.cam.ac.uk).

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METHODS
No statistical methods were used to predetermine sample size. The experiments were not randomized and the investigators were not blinded to allocation during experiments and outcome assessment.

Expression and purification of mini-Gα. The mini-Gα construct used (construct 414) was identical to mini-Gα (construct 393) described elsewhere (B.C. and G.C.T., manuscript submitted), except that one additional mutation, L63Y, was included to improve crystal quality. An N-terminal histidine tag (His6) and TEV protease cleavage site were present to facilitate purification. Mini-Gα was expressed in E. coli strain BL21(DE3)RIL upon induction with IPTG (50 μM) for 20 h at 25 °C. Cells were harvested by centrifugation and lysed by sonication in lysis buffer (40 mM HEPES pH 7.5, 100 mM NaCl, 10% glycerol, 10 mM imidazole, 5 mM MgCl2, 50 μM GDP, 1 mM PMSF, 2.5 μM Pepstatin-A, 10 μM Leupeptin, 50 μg/ml DNase I, 100 μg/ml lysozyme, 100 μM DTT), supplemented with Complete protease inhibitors (Roche). The lysate was clarified by centrifugation and loaded onto a 10 ml Ni2+– Sepharose FF column. The column was washed with wash buffer (20 mM HEPES pH 7.5, 500 mM NaCl, 40 mM imidazole, 10% glycerol, 1 mM MgCl2, 50 μM GDP) and eluted with elution buffer (20 mM HEPES pH 7.5, 100 mM NaCl, 500 mM imidazole, 10% glycerol, 1 mM MgCl2, 50 μM GDP). TEV protease was added and the sample was dialysed overnight against dialysis buffer (20 mM HEPES pH 7.5, 100 mM NaCl, 10% glycerol, 1 mM MgCl2, 10 μM GDP). TEV protease was removed by negative purification on Ni2+-NTA resin (Qiagen). The sample was concentrated to 1.5 ml and loaded onto a Superdex-200 26/600 gel-filtration column, equilibrated with gel-filtration buffer (10 mM HEPES pH 7.5, 100 mM NaCl, 10% glycerol, 1 mM MgCl2, 1 μM GTP, 0.1 mM TCEP). Peak fractions were pooled and concentrated to 100 mg/ml. The pure protein was aliquoted, flash-frozen in liquid nitrogen, and stored at −80 °C. A typical yield was 100 mg of pure mini-Gα per litre of culture.

Expression and purification of adenosine A2AR. Wild-type human adenosine A2AR were prepared using the flashBAC ULTRA system (Oxford Expression Technologies). Trichoplusia ni cells were grown in suspension in ESP921 media (Expression Systems) to a density of 3 × 10^5 cells/ml, infected with A2AR baculovirus and incubated for 72 h. Cells were harvested and membranes prepared by two ultracentrifugation steps in 20 mM HEPES pH 7.5, 1 mM EDTA, 1 mM PMSF, NECA (100 μM), NaCl (300 mM), PMSF (1 mM) and Complete protease inhibitors (Roche) were added to the membranes, and the sample was mixed for 30 min at room temperature. Membranes from 3 l of cells were solubilized with 2% n-decyl-β-D-maltopyranoside (DM) on ice for 1 h. The sample was clarified by ultracentrifugation and loaded onto a 5 ml Ni2+–NTA column (Qiagen). The column was washed with wash buffer (20 mM HEPES pH 7.5, 500 mM NaCl, 10% glycerol, 80 mM imidazole, 100 μM NECA, 0.15% DM), and eluted with elution buffer (20 mM HEPES pH 7.5, 100 mM NaCl, 10% glycerol, 300 mM imidazole, 100 μM NECA, 0.15% DM). The eluate was concentrated to 50 kDa using a 50 kDa cut-off Amicon centrifugal ultrafiltration unit (Millipore), and exchanged into dialysis buffer (20 mM HEPES pH 7.5, 100 mM NaCl, 10% glycerol, 1 mM MgCl2, 100 μM NaOAc pH 5.5, 9.5% PEG 2000 MME (in the absence of CHS). Crystals were cryo-protected in mother liquor supplemented with 30% PEG 400 and flash frozen in liquid nitrogen.

Data collection, processing and refinement. Diffraction data were collected at the European Synchrotron Radiation Facility on beamline ID23-2 with a Pilatus 2M detector, using a 6 μm × 6 μm microfocus beam (0.8729 Å wavelength). Data were indexed using either standard or helical collection modes. Data from two crystals were used for structure solution. Data were processed using MOSFLM25 and AIMLESS26. The structure was solved by molecular replacement with PHASER27 using the structures of the thermostabilized A2AR (PDB ID: 2YDV) and the Gα, GTase domain (residues 40–59 and 205–394) from the β3AR–G, complex (PDB ID: 3SNE) as search models. Model refinement and rebuilding were performed using REFMAC28 and COOT29.

Competition binding assay. FreeStyle HEK293-F cells transiently expressing wild-type A2AR were resuspended in either assay buffer A (25 mM HEPES, pH 7.5, 100 mM KCl, 1 mM MgCl2), assay buffer B (25 mM HEPES, pH 7.5, 100 mM NaCl, 1 mM MgCl2), or assay buffer C (25 mM HEPES, pH 7.5, 500 mM NaCl, 1 mM MgCl2), and were lysed by 10 passages through a 26-gauge needle. Purified binding partners were buffer-exchanged to the respective buffer before being added to the membranes at a final concentration of 25 μM. The mixture was aliquoted and NECA was added (0 to 1 mM final concentration, prepared in assay buffers containing 1 U/ml apyrase). The samples were incubated for 90 min at 22 °C, 1H-ZM241385 was added at its apparent Kd (2.5 μM) and allowed to bind for a further 90 min at 22 °C. Non-specific binding was determined in the presence of 100 μM of ZM241385. Receptor-bound and free radioligand were separated by filtration through 96-well GF/B filter plates (pre-soaked with 0.1% polyethyleneimine), and washed three times with the appropriate buffer. Plates were dried and radioactivity was quantified by liquid scintillation counting using a Tri-Carb 2910 TR (Perkin Elmer). Data were analysed by non-linear regression using GraphPad Prism software. The Kd for NECA binding was derived from one-site fit Kd analysis. Data from at least three independent experiments, each performed in duplicate, were analysed using an unpaired two-tailed t-test for statistical significance.

Thermostability assay. Membranes from Trichoplusia ni cells expressing wild-type human A2AR were resuspended in T60 buffer (25 mM HEPES pH 7.5, 100 mM NaCl, 1 mM MgCl2) and homogenized by ten passages through a 26-gauge needle. Binding partner was added at a final concentration of 25 μM. 3H-NECA and unlabelled NECA were mixed in a molar ratio of 1:5 and added to the membranes to give a final concentration of 1 μM (approximately tenfold above the apparent Kd). The samples were incubated at room temperature for 1 h, then chilled on ice for 30 min. DDM, DM or OG were added to a final concentration of 0.1%, 0.13% or 0.8%, respectively, and samples were incubated on ice for 1 h. Cell debris and insoluble material were removed by centrifugation at 5 min at 20,000g and the supernatant was aliquoted into PCR strips. Samples were heated to the desired temperature for exactly 30 min, then quenched on ice for 30 min. Samples (50 μl) were loaded onto gel-filtration resin packed into a 96-well filter plate (Millipore), which was centrifuged to separate receptor-bound from free radioligand. Non-specific binding was determined in the presence of 200 μM unlabelled NECA. Radioactivity was quantified by liquid scintillation counting using a MicroBeta TriLux scintillation counter (PerkinElmer). Data were analysed by nonlinear regression using GraphPad Prism software. Apparent Tm values were derived from sigmoidal dose–response analysis. Results represent the mean ± s.e.m. of two independent experiments, performed in duplicate.

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Extended Data Figure 1 | Pharmacological analyses of A2A R–mini-G complex. Competition assays were performed on A2A R expressed in HEK293 cell membranes with the agonist NECA competing for the binding of radiolabelled inverse agonist ³H-ZM241385. Experiments were performed in the presence of either 100 mM KCl (a, b), 100 mM NaCl (c, d) or 500 mM NaCl (e, f) to confirm the similar behaviour of mini-Gs with heterotrimeric Gs with nanobody Nb35 for stabilization of the complex. Results are summarized in the Table (g). All error bars represent the s.e.m. for at least three independent experiments performed in duplicate. Comparisons of data in b, d and f were performed using an unpaired t-test with significance denoted by asterisks: ***P < 0.0001; **P < 0.01; *P < 0.1; not significant (NS) P > 0.1.
Extended Data Figure 2 | Thermostability of detergent-solubilized 
\( ^3 \)H-NECA-bound \( \text{A}_{2A} \text{R} \) in the presence or absence of mini-Gs.

Unpurified \( \text{A}_{2A} \text{R} \) was solubilized in detergent at the following concentrations: \( \text{a} \), DDM, 0.1%; \( \text{b} \), DM, 0.13%; \( \text{c} \), OG, 0.8%. Samples were heated for 30 min, quenched on ice and the amount of \( ^3 \)H-NECA bound determined. Data were analysed by nonlinear regression and apparent \( T_m \) values (transition temperature where 50% of the receptor is inactive) were determined from analysis of the sigmoidal dose–response curves fitted (\( \text{d} \)). Results represent the mean ± s.e.m. of two independent experiments, performed in duplicate.
Extended Data Figure 3 | Omit maps for NECA and GDP.

a–f, Orthogonal views of omit map difference density for NECA in A2AR chain A (a, b), NECA in A2AR chain B (c, d) and GDP in mini-G protein chain C (e, f). The contour level is 2.5σ in panels a–d and 3.0σ in panels e and f. Figures were made using CCP4mg\textsuperscript{30}.
Extended Data Figure 4 | Electron density for the interface region of the A2AR–mini-Gs complex. The backbones of A2AR and mini-Gs are shown in cartoon representation in light blue and magenta respectively. Side chains are shown in stick representation (carbon, light blue; oxygen, red; nitrogen, deep blue). The electron density of the final $2F_o - F_c$ map is shown contoured at 1.2σ. For clarity, transmembrane helices H5 and H6 and the corresponding electron density have been omitted. a, View showing the interaction between the C-terminal helix of mini-Gs and the CL2 loop of A2AR. b, View showing the interactions between side chains of the C-terminal helix of mini-Gs and three Arg residues of A2AR.
Extended Data Figure 5 | Alignment of mini-Gs with GNAS2. Comparison of amino acid residues in mini-Gs (chains C and D) within 3.9 Å of A2AR (green) in the A2AR–mini-Gs structure and the amino acid residues in bovine GNAS2 (P04896) within 3.9 Å of β2AR in the β2AR–Gs structure (turquoise). The CGN system is used for reference.
Extended Data Figure 6 | Alignment of β2AR and A2A R amino acid sequences. adrb2_human, human β2-adrenergic receptor; AA2AR_human, human adenosine A2A receptor; AA2AR chain A, chain A of the crystallized A2AAR mini-Gs structure; AA2AR chain B, chain B of the crystallized A2AAR mini-Gs structure. Residues in the receptors that are within 3.9 Å of either GαL in the β2AR–Gs complex or mini-Gs in the A2AAR–mini-Gs complex are highlighted in turquoise or green, respectively. Key Ballesteros–Weinstein numbers are shown in blue and mutations in the crystallized A2AAR to facilitate purification and crystallization are shown in red. Grey bars indicate the positions of α-helices in the β2AR–Gs structure, whereas red bars represent these regions in the A2AAR–mini-Gs structure; where there is a discrepancy in helix length between chain A and B of A2AAR, the bar is coloured pink.
Extended Data Figure 7 | A conserved hydrophobic binding pocket at the receptor–Go subunit interface. The A2AR–mini-Gs complex was aligned to the β2AR–Gs complex via the receptors: A2AR, green; β2AR, turquoise; mini-Gs, purple; Goα, grey.
Extended Data Figure 8 | Comparison between receptor-bound mini-G<sub>s</sub> and G<sub>α</sub><sub>sα</sub>. a–c, Three different views of an alignment of mini-G<sub>s</sub> (chain C, purple) bound to A<sub>2a</sub>R with the GTPase domain of G<sub>α</sub><sub>sα</sub> (grey) bound to β<sub>2</sub>AR. GDP bound to mini-G<sub>s</sub> is depicted as a space filling model (carbon, yellow; oxygen, red; nitrogen, blue; phosphorus, orange). The α<sub>5</sub> helix that interacts with the receptors is labelled.
Extended Data Figure 9 | Comparison of the NECA binding site in the active-intermediate state compared to the mini-\(G_s\)-bound state. The structure of NECA-bound \(A_{2A}R\) (grey cartoon, with the carbon atoms of NECA also in grey) in the active-intermediate state was aligned with the structure of the NECA-bound \(A_{2A}R\)-mini-\(G_s\) complex (rainbow colouration, with the carbon atoms of NECA in green). Key amino acid residues for both receptors are depicted (sticks; carbon atoms in the same colour as the respective receptor) that form hydrogen bonds (red dashed line) with either NECA or the associated water network (red spheres). Note that the water molecules depicted are from only the NECA-bound \(A_{2A}R\) structure in the active-intermediate state, because the resolution of the \(A_{2A}R\)-mini-\(G_s\) structure was insufficient to identify water molecules. Carbonyl oxygens are denoted by 'co' after the residue name.
Extended Data Table 1 | Data collection and refinement statistics

| **Data collection** |  |
|---------------------|---|
| Space group         | P 2₁ 2₁ 2₁ |
| Cell dimensions $a$, $b$, $c$ (Å) | 90.6, 111.8, 161.3 |
| Resolution (Å) $^1$ | 40.3-3.4 (3.49-3.40) |
| $R_{merge}$         | 0.173 (0.747) |
| $I/σI$              | 3.6 (1.2) |
| Completeness (%)    | 90.6 (78.5) |
| Redundancy          | 2.6 (2.4) |

| **Refinement** |  |
|----------------|---|
| Resolution (Å) | 40.3-3.4 |
| No. reflections | 19788 |
| $R_{work}/R_{free}$ (%) | 28.4/31.5 |
| No. atoms       | 7359 |
| Protein         | 7248 |
| Ligand/detergent/nucleotide | 44/40/27 |
| Water           | 0 |
| B-factors (Å$^2$) |  |
| Protein         | 79.9 |
| Ligand/detergent/nucleotide | 67.9/98.6/69.0 |
| R.M.S.D.        |  |
| Bond lengths (Å) | 0.008 |
| Bond angles (°) | 1.15 |

$^1$Values in parentheses are for the highest resolution shell.
Erratum: Structure of the adenosine A$_{2A}$ receptor bound to an engineered G protein

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In this Letter, author B.C. (byronc@mrc-lmb.cam.ac.uk) should have also been included as a corresponding author; this has been corrected online.