Supporting Information

Sato et al. 10.1073/pnas.0809974106

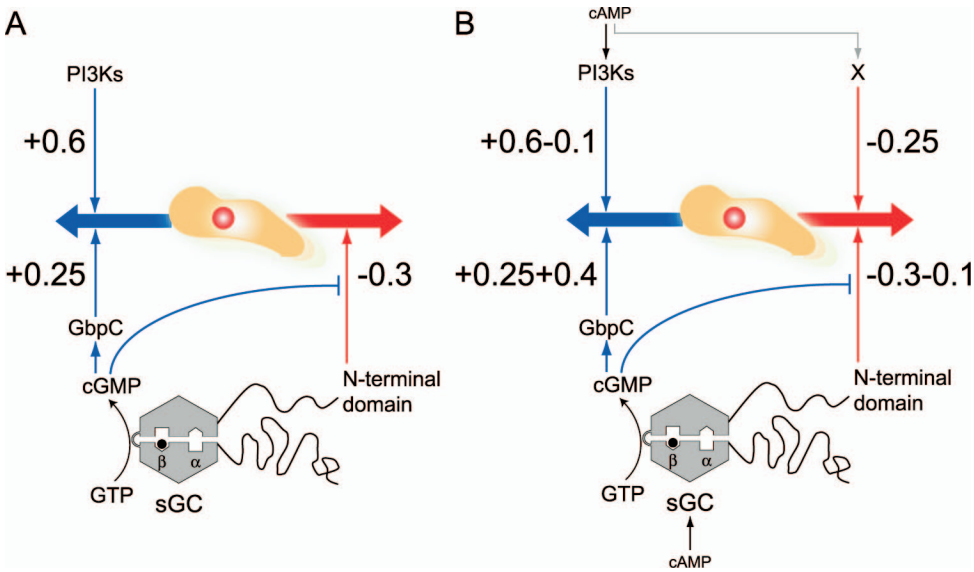
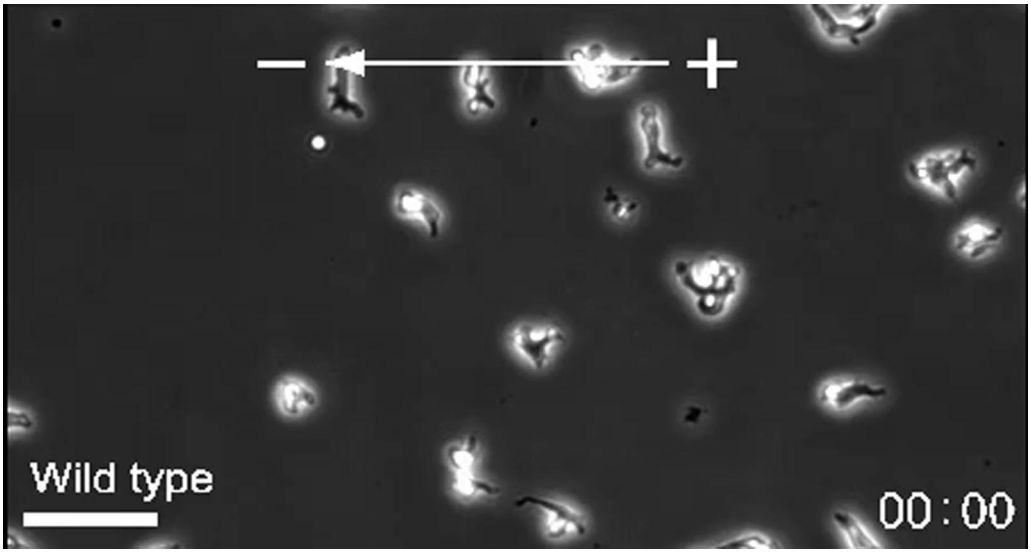
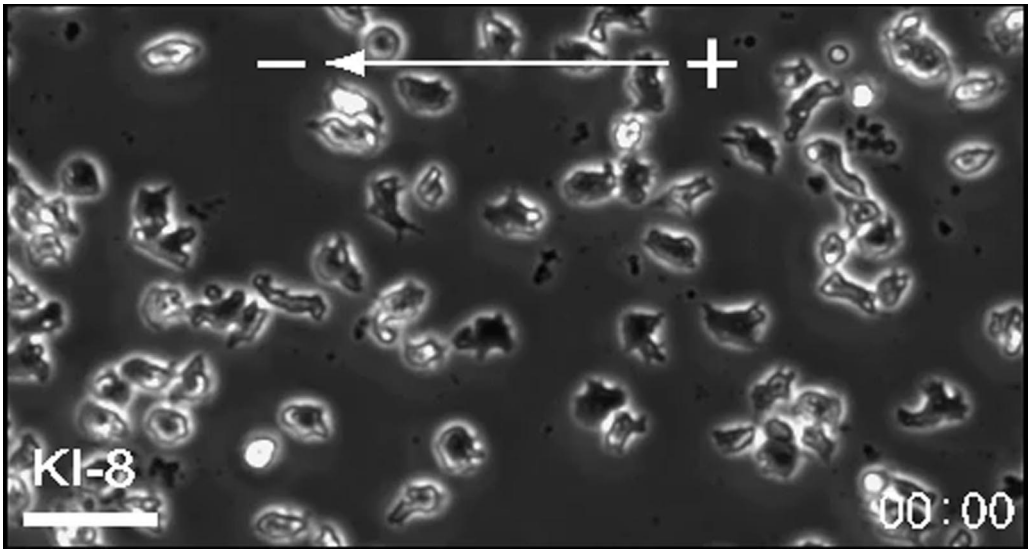


Fig. S1. Multiple signaling pathways for electrotaxis in *Dictyostelium* cells. PI3Ks and cGMP-GbpC pathways mediate cathode-directed migration, whereas the N-terminal domain of sGC and an unidentified pathway (X) mediate anode-directed migration. Multiple-signaling pathways in the absence of cAMP (*A*) and in the presence of cAMP (*B*). Numbers represent relative strength of bias for individual-signaling pathways, where positive and negative values indicate directedness for migration toward the cathode and anode, respectively. Because PI3Ks and GCase depend on extracellular cAMP stimulation in chemotaxis, the relative strength of bias for individual signaling pathways are modulated by cAMP stimulation.



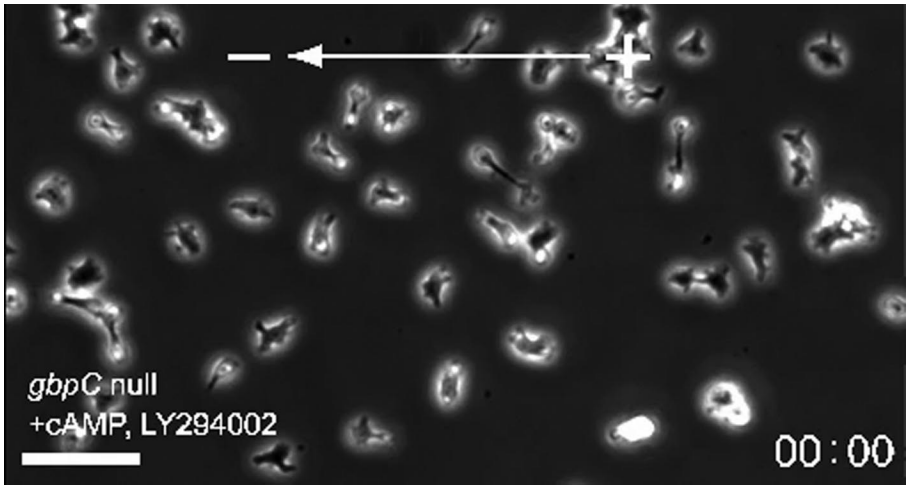
Movie S1. Cathode-directed migration of WT *Dictyostelium* cells in a dcEF. Starved cells migrated toward the cathode (left). Observation time was 20 min over 5-sec intervals. Electric field (EF) = 10 V·cm^{-1}. (Scale bar, 50 μm.)

Movie S1 (MOV)



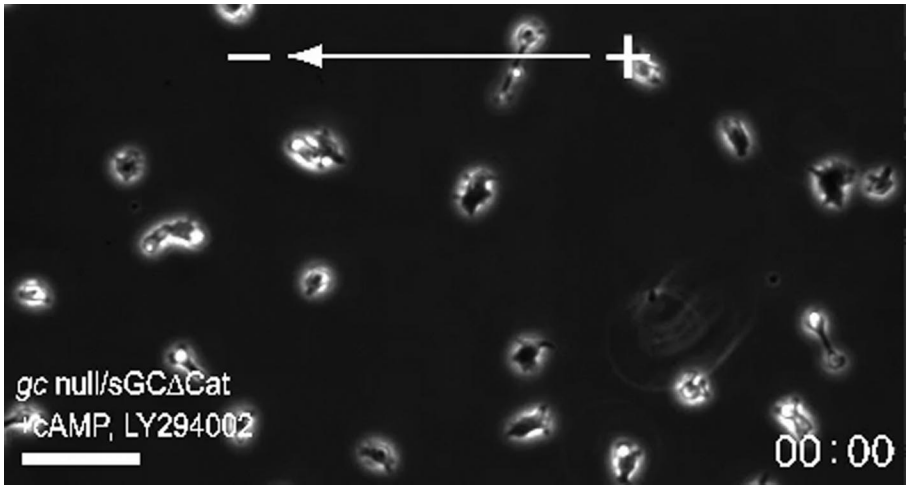
Movie S2. Anode-directed migration of KI-8 mutant cells in a dcEF. KI-8 cells, which have no guanylyl cyclase activity, migrated toward the anode with a slower migration velocity than that of WT cells. Observation time was 20 min over 5-sec intervals. EF = 10 V·cm^{-1}. (Scale bar, 50 μm.)

Movie S2 (MOV)



Movie S3. Anode-directed migration in cells lacking cGMP-binding protein C (*gbpC*$^{-}$) in a dcEF. Simultaneous suppression of both cGMP-binding proteins and PI3K activities caused reversal of cell migration direction. Observation time was 20 min over 5-sec intervals. EF = 10 V·cm^{-1}. (Scale bar, 50 μm.)

Movie S3 (MOV)



Movie S4. Anode-directed migration of *gc*-null/sGCΔCat cells in a dcEF. Absence of intracellular cGMP and suppression of PI3K activity caused continuous reversal migration. Observation time was 20 min over 5-sec intervals. EF = 10 V·cm^{-1}. (Scale bar, 50 μm.)

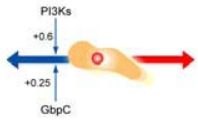
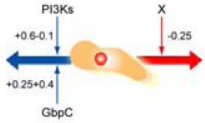
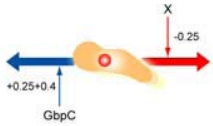
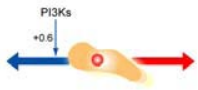
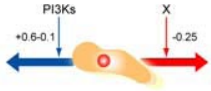
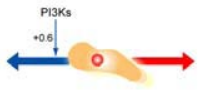
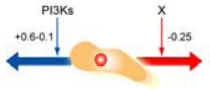
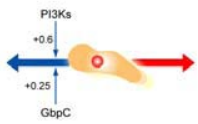
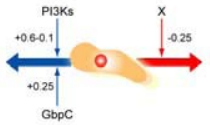
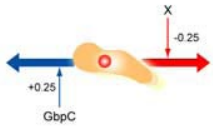
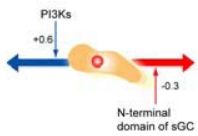
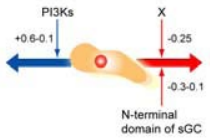
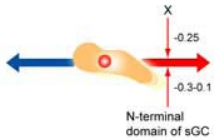
Movie S4 (MOV)

Table S1. Cell migration properties in a dcEF

Cell type	EF stimulation (10 V·cm^{-1})	PI3K inhibitor (60 μM LY294002)	cAMP (1 μM)	No. of cells analyzed	Directedness	Migration velocity, μm/min	Path linearity	Asymmetric index (cathode:anode)
WT	−	−	−	157	0.01 ± 0.06 (Random)	11.8 ± 0.4	0.4 ± 0.02	0.49:0.51
WT	+	−	−	119	0.88 ± 0.02 (Cathode)	14.5 ± 0.4	0.61 ± 0.01	0.98:0.02
WT	+	−	+	43	0.95 ± 0.01 (Cathode)	16.5 ± 0.6	0.64 ± 0.02	1:0
WT	−	+	−	137	0.05 ± 0.06 (Random)	2.8 ± 0.2	0.14 ± 0.01	0.5:0.5
WT	−	+	+	119	0.06 ± 0.07 (Random)	7.8 ± 0.2	0.39 ± 0.02	0.49:0.51
WT	+	+	−	148	0.53 ± 0.05 (Cathode)	4.6 ± 0.2	0.26 ± 0.02	0.8:0.2
WT	+	+	+	99	0.36 ± 0.06 (Cathode)	14 ± 0.4	0.54 ± 0.02	0.75:0.25
gbpA^{-}/*gbp*$^{-}$	+	−	−	110	0.9 ± 0.02 (Cathode)	13.7 ± 0.3	0.67 ± 0.01	0.99:0.01
KI-5	−	−	−	107	0.11 ± 0.07 (Random)	22 ± 0.4	0.39 ± 0.02	0.59:0.41
KI-5	+	−	−	103	0.84 ± 0.03 (Cathode)	25 ± 0.5	0.64 ± 0.01	0.96:0.04
KI-8	−	−	−	132	−0.06 ± 0.06 (Random)	5.9 ± 0.2	0.38 ± 0.02	0.47:0.53
KI-8	+	−	−	142	−0.62 ± 0.04 (Anode)	8.4 ± 0.3	0.5 ± 0.02	0.13:0.87
KI-10	−	−	−	125	−0.04 ± 0.06 (Random)	25.8 ± 0.3	0.5 ± 0.02	0.49:0.51
KI-10	+	−	−	116	0.04 ± 0.07 (Random)	26.4 ± 0.6	0.47 ± 0.02	0.53:0.47
gc-null	+	−	−	111	0.63 ± 0.05 (Cathode)	14.1 ± 0.4	0.45 ± 0.02	0.89:0.11
gc-null	+	+	+	138	−0.17 ± 0.06 (Anode)	8.4 ± 0.2	0.42 ± 0.01	0.39:0.61
gc-null/sGCΔN	+	−	−	133	0.83 ± 0.03 (Cathode)	9.4 ± 0.4	0.6 ± 0.01	0.95:0.05
gc-null/sGCΔN	+	−	+	136	0.45 ± 0.05 (Cathode)	9.3 ± 0.4	0.54 ± 0.02	0.77:0.23
gc-null/sGCΔN	+	+	+	114	0.01 ± 0.06 (Random)	7.0 ± 0.3	0.53 ± 0.02	0.53:0.47
gc-null/sGCΔCat	+	−	−	105	0.32 ± 0.07 (Cathode)	12.5 ± 0.4	0.5 ± 0.02	0.69:0.31
gc-null/sGCΔCat	+	−	+	121	−0.26 ± 0.06 (Anode)	12.4 ± 0.3	0.62 ± 0.02	0.34:0.66
gc-null/sGCΔCat	+	+	+	113	−0.67 ± 0.04 (Anode)	10.6 ± 0.2	0.73 ± 0.01	0.09:0.91
gbpC-null	+	−	−	139	0.61 ± 0.04 (Cathode)	13.3 ± 0.2	0.47 ± 0.02	0.87:0.13
gbpC-null	+	+	+	190	−0.36 ± 0.05 (Anode)	7.8 ± 0.1	0.39 ± 0.01	0.28:0.72

Data are presented as mean ± SEM. Definition of each parameter is given in Sato MJ, et al. (2007) Input-output relationship in galvanotactic response of Dictyostelium cells. *BioSystems* 88:261–272. Data for each cell type was collected from 2–11 independent experiments.

Table S2. Directedness estimated by multiple-signaling pathway model for electrotaxis

Cell type	-	+cAMP	+cAMP, LY294002
WT	Cathode (0.88)*	Cathode (0.95)	Cathode (0.36)
	 (0.85)†	 (0.9)	 (0.4)
gc-null	Cathode (0.63)	ND‡	Anode (-0.17)
	 (0.6)	 (0.25)	 (-0.25)
gbpC-null	Cathode (0.61)	ND	Anode (-0.36)
	 (0.6)	 (0.25)	 (-0.25)
gc-null/ sGCΔN	Cathode (0.83)	Cathode (0.45)	Random (0.01)
	 (0.85)	 (0.5)§	 (0)
gc-null/ sGCΔCat	Cathode (0.32)	Anode (-0.26)	Anode (-0.67)
	 (0.3)	 (-0.15)	 (-0.65)

*Directedness measured experimentally.
†Directedness calculated from hypothetical strength of bias.
‡Not determined experimentally in this condition.
§Here, we assume that cAMP stimulus fails to activate the GbpC activity in the absence of the N-terminal domain of sGC [Veltman DM, Roelofs J, Engel R, Visser AJ, Van Haastert PJ (2005) Activation of soluble guanylyl cyclase at the leading edge during Dictyostelium chemotaxis. *Mol Biol Cell* 16:976–983]. In this table, left and right sides indicate cathode and anode, respectively.