letters to nature

the wild-type subcomplex 52 ATP s⁻¹, and the native F₁-ATPase 39 ATP s⁻¹. Rabbit skeletal actin (30 µM) was incubated with 150 µM biotin-PEAC5maleimide in 100 mM KCl, 1 mM MgCl₂, 10 mM MOPS-KOH (pH 7.0) and 0.3 mM NaN₃ at room temperature for 2 h. The actin was depolymerized in 2 mM MOPS-KOH (pH 7.0), 0.2 mM CaCl₂ and 2 mM ATP. Residual biotin was removed on a Sephadex G-25 column. Biotinylated actin (5 µM) was polymerized in 10 mM 2-(cyclohexylamino)ethanesulphonic acid-KOH (pH 8.8), 100 mM KCl, 1 mM MgCl₂ and 5 µM phalloidin-tetramethylrhodamine B isothiocyanate conjugate (Fluka) overnight at 4°C, and crosslinked with 500 µM disuccinimidyl suberate (Pierce) at room temperature for 2 h. The reaction was quenched with 50 mM Tris-HCl (pH 8.8).

Immobilization of proteins. A flow cell for microscopic observation was constructed from a bottom coverslip ($24 \times 36 \text{ mm}^2$; Matsunami) coated with nitrocellulose and a top coverslip $(18 \times 18 \text{ mm}^2)$, separated by two greased strips of Parafilm cover sheet. 0.6-1.2 µM of horseradish peroxidaseconjugated Ni-NTA (Qiagen) was introduced into the flow cell and allowed to adhere to the glass surface for 2 min. The cell was washed with buffer A (10 mg ml⁻¹ BSA, 10 mM MOPS-KOH (pH 7.0), 50 mM KCl, 4 mM MgCl₂). Infusion and washing were repeated as follows: infusion of 10-100 nM biotinylated $\alpha_3\beta_3\gamma$ subcomplex in buffer A (5 min), washing with buffer A, infusion of 180 nM streptavidin (Sigma) in buffer A (2 min), washing with buffer A, and infusion of 100 nM biotinylated fluorescent actin filaments in buffer A (5-15 min). The last wash was carried out with 0.5% 2-mercaptoethanol and an oxygen-scavenger system²² in buffer A containing, where indicated, 2 mM ATP or 10 mM NaN₃. Observation started within 1 min of the beginning of the last washing. The actin filaments did not bind to the glass plate without the biotinylated subcomplexes. Also, the binding was dependent on streptavidin. In a control experiment, His-tagged subcomplexes fluorescently labelled at γ -Cys107 were fixed on a Ni-NTA surface and extensively washed with buffer A; subsequent washings with buffer A containing 50 mM imidazole (pH 7.4) removed >85% of the fluorescence. These results ensure that the actin filaments were attached to the biotinylated $\alpha_3\beta_3\gamma$ subcomplexes which were fixed to the glass surface through the histidine tags.

Observation of rotation. Actin filaments were observed under an epifluorescence microscope (Diaphot TMD, Nikon) with excitation and emission wavelengths at 546 nm and 560-620 nm, respectively. Images were taken with a CCD camera (Dage MTI) attached to an image intensifier (KS-1381, Videoscope), recorded on an 8-mm video tape, and analysed with a digital image processor (DIPS-C2000, Hamamatsu Photonics)^{2,23}. The frictional torque for the propeller rotation is given²⁴, in the simplest approximation, by $(\pi/3)\omega\eta L^3/[\ln(L/2r) - 0.447]$, where ω is the angular velocity, $\eta (10^{-3}$ N s m⁻²) the viscosity of the medium, L the length of actin filament, and r (5 nm) the radius of the filament. For the rotation around one end of the filament, the torque is four times the above value. These values are actually underestimated, because the viscous drag near the glass surface is higher (up to \sim 3-fold²⁴ if all of the filament lies at a height of (5 + 8) nm from the glass surface, 5 nm being the filament radius and 8 nm the height⁸ of $\alpha_3\beta_3$) and because possible contact with the surface would produce additional friction.

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erratum

Spatio-temporal frequency domains and their relation to cvtochrome oxidase staining in cat visual cortex

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Nature 385, 529-533 (1997)

In this Letter, an editing error in the Nature office led to a misleading first sentence in the last paragraph. The complete paragraph should read as follows:

Tracing studies have shown that blobs in cat visual cortex are specifically connected to other visual cortical areas^{29,30} and that they receive a strong input from Y-cells in the lateral geniculate nucleus¹¹. Furthermore, we have preliminary evidence that low spatial frequency domains, like blobs, receive a stronger input from geniculate Y-cells³ which would be consistent with the preference for low spatial and high temporal frequencies revealed here. The blob and interblob regions of cat visual cortex, like their counterparts in primate visual cortex, appear to be compartments of parallel pathways that are specialized for analysing different attributes of the visual scene.

correction

Transmission dynamics and epidemiology of BSE in British cattle

R. M. Anderson, C. A. Donnelly, N. M. Ferguson, M. E. J. Woolhouse, C. J. Watt, H. J. Udy, S. MaWhinney, S. P. Dunstan, T. R. E. Southwood, J. W. Wilesmith, J. B. M. Ryan, L. J. Hoinville, J. E. Hillerton, A. R. Austin & G. A. H. Wells

Nature 382, 779-788 (1996).

..... In Table 2 of this Letter, the reported number of cases saved for policy 9 was erroneously given for 1996 to 2001 rather than for 1997 to 2001. The number should be 584 rather than 797. Thus, the number of cases saved in policies 11-14 should be reduced by 213. Also, the culling policy description for policy 6 should begin 'As 5)' rather than 'As 15)'.

An error caused Figs 1d and e in this article to be transposed. The legends are correct.

On page 783, in the first column in the sixth line of text 'F and G are two operators' should have been 'G and F are two operators'.

Finally, the manuscript on maternal transmission of the BSE agent in cows by J. W. Wilesmith et al. described in the paper as ref. 6, under consideration by Nature, has been withdrawn.