late Ca^{2+} and the substrates (arginine and NADPH) needed for NO synthesis but will produce ·NO or O_2^- only when oxygen is supplied by reperfusion. Intravenously administered superoxide dismutase may reduce ischaemic injury by scavenging O_2^- before it reacts with ·NO to form $ONOO^-$.

This hypothesis is consistent with current data showing the reduction of ischaemic injury by superoxide dismutase, desferrioxamine and ·OH scavengers, but it also predicts that increased amounts of nitrate, nitrite and nitrosamines should be detectable in ischaemic tissue and its venous drainage.

JOSEPH S. BECKMAN
Department of Anesthesiology, University of Alabama at Birmingham, Birmingham, Alabama 35294, USA

1. Feigl, E. O. *Nature* **331**, 490–491 (1988).
2. Blough, N. V. & Zafiriou, O. C. *Inorg. Chem.* **24**, 3502–3504 (1985).
3. Beckman, J. S., Beckman, T. W., Chen, J., Marshall, P. A. & Freeman, B. A. *Proc. natn. Acad. Sci. U.S.A.* **87**, 1620–1624 (1990).
4. Marletta, M. A., Yoon, P. S., Iyengar, R., Leaf, C. D. & Wishnok, J. S. *Biochemistry* **27**, 8706–8711 (1988).
5. Moncada, S., Palmer, R. M. J. & Higgs, E. A. *Biochem. Pharmacol.* **38**, 1709–1715 (1989).

To flow or not to flow?

SIR—Bretscher, in a recent Scientific Correspondence[1], has questioned some of the conclusions drawn in our recent articles[2,3] and renewed his advocacy of his lipid-flow hypothesis. We had shown that in a stationary reference frame, cell surface proteins labelled with small gold beads could be in either of two states: randomly diffusing or systematically transporting rearwards toward the cell centre. Because there was no detectable systematic drift of the diffusing proteins, we concluded that lipid flow does not drive the motion either of the diffusing or of the transported particles.

One question of Bretscher's arose from a misunderstanding concerning the velocity of rearwards movement of cell surface components relative to forward motion of the cell. He suggested that the apparently randomly diffusing particles were in fact moving backwards on the cell surface because the cells were moving forward at a comparable rate. In our study, however, forward motion of the cells was much slower (20–100-fold) than the rearwards transport of surface components. Hence, the absence of rearward drift of surface particles observed in the laboratory reference frame also holds for reference frames fixed to the cells. In a later study of rapidly moving cells[4], we have also observed that the diffusing particles do not display systematic transport relative to the cell even after the cell has migrated several cell lengths.

The second question Bretscher raises was whether our particles were really on the surface of the cell. The strongest evidence that they were is that 0.5-micrometre beads, which can clearly be shown to be on the upper surface of the lamellipodium by varying the focal plane of the microscope, also do not move toward the rear when they are diffusing. There are additional arguments that the viscosity of cytoplasm in the lamellipodium would not permit such rapid diffusion and we have documented similar diffusive behaviour of gold particles which were subsequently released from the membrane into the medium. Further, direct measurement of lipid labels in a photobleaching experiment has detected no rearward movement of bulk lipid even though the cells were moving forward at 40 micrometres per minute[5].

As in the case of the fabled unicorn, there may be somewhere where lipid flow can be found, but we have not seen evidence for it in motile cells.

MICHAEL SHEETZ
ELLIOT ELSON
DENNIS KUCIK
Department of Cell Biology and Physiology, Washington University School of Medicine, St Louis, Missouri 63110, USA

1. Bretscher, M. *Nature* **341**, 491–492 (1989).
2. Sheetz, M. *et al. Nature* **340**, 284–288 (1989).
3. Kucik, D. *et al. Nature* **340**, 315–317 (1989).
4. Kucik, D. *et al. J. Cell Biol.* **109**, 72a (1989).
5. Lee, J. *et al. Science* **244**, 1229–1233 (1990).

Myelination in severed nerves

SIR—There are other explanations for the interesting observation by Voyvodic[1] of increased myelination in the sympathetic nerve innervating the submandibular gland in rats, than those implied by his title "Target size regulates calibre and myelination of sympathetic axons". He says that "relative target size was increased by cutting one branch of the sympathetic nerve innervating the gland at birth", but no details are given as to which branch was cut, or how.

In 1900 Langley[2] made the surprising discovery that, after excision of the right superior cervical ganglion in the cat, the submandibular sympathetic nerve trunks on the submandibular artery did not disappear, as expected, and that one year later "there were more medullated fibres, and apparently fewer non-medullated, in the nerve strands on the artery on the right side than in those on the left — the intact side". He found no evidence for any functional reinnervation, and said that he would leave the cause of the regeneration for further investigation.

Some years ago, I followed up this unusual observation. I found that after superior cervical ganglionectomy in cats, although the non-myelinated axons had degenerated in the sympathetic trunks on the submandibular artery by day 12, some myelinated axons always persisted[3]. Subsequently, it was found[4] that some myelinated axons were present at all stages after ganglionectomy and that their numbers increased with time up to one year. Despite the drastic procedure of sectioning the post-ganglionic sympathetic trunks, together with ligature and transection of the associated external carotid artery — just proximal to the submandibular artery — some myelinated axons in the trunks on the submandibular artery always survived and in time they increased in numbers.

The source of the myelinated axons in the submandibular trunks that withstand such nearby axotomies remains a mystery. Sometimes, small axons with newly forming myelin were seen after a few weeks close to myelinated axons of more normal size, so they may have arisen from sprouting in the vicinity, alternatively *de novo* myelination may have been occurring.

Histochemical assessments of cholinesterase activity in the sympathetic nerve trunk on the submandibular artery, after post-ganglionic axotomy[5], are of interest. Normally, these sympathetic trunks show very weak acetylcholinesterase staining and somewhat stronger non-specific cholinestrase activity[6]; this is the converse of the reaction in post-ganglionic parasympathetic nerves, which show very strong acetylcholinesterase activity. After superior cervical ganglionectomy, there is an early loss of all cholinesterase staining in the submandibular sympathetic trunks, followed by a gradual increase, especially of acetylcholinesterase activity. In other words, the submandibular sympathetic trunks come histochemically to resemble parasympathetic nerves. Since these changes were initially more evident closer to the gland, it was suggested (ref. 5) that the unmyelinated axons in the trunks had arisen from collateral sprouting of cholinergic post-ganglionic parasympathetic nerves within the gland, which had then migrated in a retrograde manner down the sympathetic trunks.

Much work remains to be done to ascertain the true cause of any increase in the number of myelinated nerves in the sympathetic trunks to the submandibular glands after denervation experiments.

J. R. GARRETT
Department of Oral Pathology, The Rayne Institute, King's College School of Medicine and Dentistry, London SE5 9NU, UK

1. Voyvodic, J. T. *Nature* **342**, 430–433 (1989).
2. Langley, J. N. *J. Physiol.* **18**, 417–426 (1900).
3. Garrett, J. R. *J. R. Micr. Soc.* **86**, 15–31 (1966).
4. Garrett, J. R. & Kemplay, S.K. *J. Anat.* **124**, 99–115 (1977).
5. Garrett, J. R. *Prog. Brain Res.* **34**, 475–488 (1971).
6. Garrett, J. R. *J. R. Micr. Soc.* **85**, 135–148 (1966).