PNAS LETTER

Early characterization of excitation–contraction coupling and SLO-2 channel properties in *Caenorhabditis elegans* muscle cells

Bruno Allard[a,1]

In a recent study, Gao et al. (1) report that voltage-induced calcium release (VICR) occurs in *Caenorhabditis elegans* body wall muscles. More than twenty years ago, the excitation–contraction (EC) coupling process was explored at the cellular level using in situ voltage clamp and intracellular calcium measurements in body wall muscle cell from *C. elegans* (2). In this pioneer article, our group demonstrated that the cytoplasmic changes in intracellular calcium upon depolarization are fully dependent on nifedipine-sensitive calcium entry through EGL-19. Calcium transients activated by depolarization and voltage-gated calcium currents were indeed shown to have the same potential value of activation and bell-shaped voltage dependence. Hence, in response to high depolarization values (+50 mV), for which the calcium entry through voltage-gated Ca^{2+} channels was drastically decreased, we showed that the calcium transients were also strongly decreased. If VICR exists, one would have expected that robust intracellular calcium changes occur in response to this elevated depolarization. In that context, our data missed by Gao et al. (1), although not disputing the existence of physical coupling between EGL-19 and UNC-68, question the physiological relevance of VICR in body wall muscle function that the authors describe in their paper. Incidentally, Gao et al. (1), by showing that mutations (*egl-19(ΔVTTL)* and *shn-1*) disrupting VICR do not affect locomotion, admit that blocking VICR has "relatively modest effects," rather no significant effects on EC coupling regarding the statistical tests provided by the authors. The fact that locomotion is not affected by VICR deficiency also questions the assumption of the authors that VICR in *C. elegans* promotes excitation–repolarization coupling by promoting SLO-2 activation. Along this line, SLO-2 expressed in *Xenopus* oocytes was shown to be calcium- and chloride- but also voltage-activated (3). Our group confirmed these data using in situ patch clamp technique on body wall muscle cells from *C. elegans* (4). By omitting to consider this voltage gating, Gao et al. (1) deduced from their experiments, consisting in preventing external calcium entry, that the observed reduction but not elimination in Ik_{hiCl} is due to partial activation of SLO-2 by VICR, whereas this remaining activation of SLO-2 could be provoked by depolarization. Additionally, removing external calcium, not compensated by other divalent cations, is known to induce surface charge effects that shift the voltage dependence of voltage-gated channels toward negative potentials. Finally, we also demonstrated that a small conductance calcium-activated potassium channel was present in body wall muscle cells of *C. elegans* (4). It is thus puzzling that preventing external calcium entry had no effect on muscle cells from *slo-2* mutants.

Author affiliations: [a]Physiopathologie et Génétique du Neurone et du Muscle, Université de Lyon, Université Claude Bernard Lyon 1, CNRS UMR 5261, INSERM U1315, Faculté de Médecine Rockefeller, Lyon 69008, France

Author contributions: B.A. wrote the paper.

The author declares no competing interest.

[1]Email: bruno.allard@univ-lyon1.fr.

Published September 16, 2024.

1. L. Gao, E. Ardiel, S. Nurrish, J. M. Kaplan, Voltage-induced calcium release in *Caenorhabditis elegans* body muscles. *Proc. Natl. Acad. Sci. U.S.A.* **121**, e2317753121 (2024).
2. M. Jospin, V. Jacquemond, M.-C. Mariol, L. Ségalat, B. Allard, The L-type voltage-dependent Ca^{2+} channel EGL-19 controls body wall muscle function in *Caenorhabditis elegans*. *J. Cell Biol.* **159**, 337–348 (2002).
3. A. Yuan *et al.*, SLO-2, a K^+ channel with an unusual Cl^- dependence. *Nat. Neurosci.* **3**, 771–779 (2000).
4. M. Jospin, M.-C. Mariol, L. Ségalat, B. Allard, Characterization of K^+ currents using an *in situ* patch clamp technique in body wall muscle cells from *Caenorhabditis elegans*. *J. Physiol.* **544**, 373–384 (2000).