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Comment

Estimating the contribution of lightning to microbial evolution: Guidance from the Drake equation
Comment on "Lightning-triggered electroporation and electrofusion as possible contributors to natural horizontal gene transfer" by Tadej Kotnik

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The possibility that lightning-mediated horizontal gene transfer (HGT) has contributed to evolution has been raised before [1–5], but not vigorously pursued. Kotnik now thoughtfully argues for doing so [6]. In addition to applauding his presentation, I suggest that estimating the significance of lightning's contribution to HGT in microbial evolution is fundamentally a quantitative issue. For this reason aspects of electroporation (EP) and the Drake equation are briefly discussed.

Fig. 1 is based on experimental exposure conditions, mainly for mammalian cells, and indicates both Supra-EP and Conventional EP [7]. The former involves creation of about 100-fold more pores during nsPEF (nanosecond pulsed electric fields) than conventional EP (pulse duration greater than about 1 μs), but nsPEF pores are mostly small. Lightning-induced electric fields in the sea and soil lie within this strength-duration space, with smaller bacteria requiring ten-fold larger strengths for pulse durations greater than ~ 1 μs.

The most relevant feature of lightning is its generation of an electric current density, J, in sea or soil. The spatially distributed J leads to electric fields that can electroporate prokaryote membranes, with these field pulse strengths, durations and repetitions defining three hemispherical zones [5,6]: lethal, HGT possible and no EP effect. Here discussion focuses on DNA delivery by electroporation without electrofusion. This seems to be the more general possibility, requiring neither bacterial protoplasts nor membrane contact.

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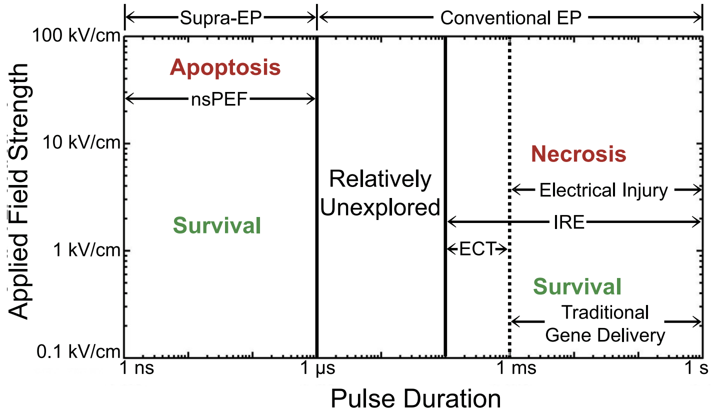

Fig. 1. Approximate map of cell electroporation pulse strength-duration space.

Experiments with laboratory electric fields have yielded several positive results [3–5]. While encouraging, future experiments could use lightning current waveforms [8] for creating electroporating pulses, initially assuming that lightning has not changed significantly over evolutionary time.

The first reports of EP are usually associated with Stämpfli and co-workers, for both irreversible [9] and reversible EP [10], but Rubinsky notes there are earlier hints of EP [11], one in 1754. As now used or studied EP field strengths span three orders of magnitude (0.1 to 100 kV/cm) and nine orders of duration (1 ns to 1 s).

Actual waveforms and multiple lightning strokes may be important. Consider electroporation-mediated HGT between two living cells. Within sea water a solubilized charged molecule moves electrophoretically without significant steric hindrance or temporary binding/unbinding to particles. Two in vitro experiments support this process, with solubilized DNA moving electrophoretically within the extracellular medium into electroporated cells directly through pores [12,13]. Within soil, however, small particles and tethered macromolecules can be present, so that released DNA is more likely to bind, perhaps temporarily, before reaching a recipient cell [3]. A related situation occurs in widely used laboratory protocols, wherein DNA is allowed to bind to the membrane before pulsing, and afterward DNA moves slowly into the cell [14–16]. Dead bacteria may provide a plasmid reservoir [5]. In soil DNA released from dead cells may remain localized until degraded, but in the sea DNA would spread by diffusion and convection. Lightning-mediated HGT requirements may therefore differ for sea and soil.

In addition to basic biophysical mechanisms of EP and DNA transport, large scale considerations should include global lightning, and the evolutionary time frame when electrical HGT would have been important. Specifically, an approach that is guided by the Drake equation may help identify the processes important to lightning-mediated HGT. This equation was proposed for a rather different evolutionary problem: Estimating the present number of extra-terrestrial civilizations with communication capability relevant to a search for extra-terrestrial intelligence (SETI) [17–22].

As emphasized recently by Dominik and Zarnecki [22] "Our ignorance is most famously quantified by the Drake equation

$$N = R_* f_p n_p f_l f_i f_c L.\text{"} \tag{1}$$

Eq. (1) "... describes the number of civilizations, N, that are detectable by means of electromagnetic emissions (more particularly, radio signals) as a product of various factors. R_* is the formation rate of suitable stars, f_p is the fraction with planetary systems, n_p is the number of planets per such system with conditions suitable for life, f_l is the fraction on which life actually develops, f_i is the fraction life-bearing planets on which intelligent life emerges, f_c is the fraction of emerged civilizations that develop technologies for propagating detectable signals, and L is the time span over which these civilizations disseminate such signals" [22].

This estimate is basically an average rate, R_*, a chain of probabilities (fractions) and one number that can be multiplied to create a single parameter, f_s [18], and a lifetime (L). It is often emphasized that the Drake equation is a way of noting what parameters are important, and this highlights the source of uncertainties. Consistent with major uncertainty, predicted values of N vary greatly [17–22].

For lightning-mediated HGT a similar predicted quantity could be the total number of lightning-mediated HGT events deemed significant during an appropriate evolutionary time frame. Quantitative estimation for the significance of lightning to HGT over evolutionary times has similar large uncertainties. For example, consider an estimate

$$N_{\mathrm{HGT}} = R_{\mathrm{LST}} n_{\mathrm{BAC}} V_{\mathrm{EPZ}} f_{\mathrm{EPT}} f_{\mathrm{SIG}} L_{\mathrm{HGT}}. \tag{2}$$

Here N_{HGT} = total number of evolutionarily significant changes due to lightning, $R_{\mathrm{LST}} = 10^{18}$ flashes/Gyr is the rate of lightning strokes on earth [23], $n_{\mathrm{BAC}} = 10^{15}$ bacteria/m^3 is the bacterial concentration (number density) in soil [24], $V_{\mathrm{EPZ}} = 1\ \mathrm{m}^3$ is the volume of the zone with successful EP, $f_{\mathrm{EPT}} = 10^{-2}$ is the fraction of successfully electroporated bacteria, and $f_{\mathrm{SIG}} = 1$ is a test value for the fraction of porated cells that experience evolutionarily significant HGT. Finally, note that $L_{\mathrm{HGT}} = 4$ Gyr is the time over which evolutionarily significant changes were created (an accumulation time, not a lifetime). Using the test value of one for f_{SIG} gives $N_{\mathrm{HGT}} = 10^{31}$, a huge number. This shows where a lot of uncertainty resides. While this probability appears very difficult to estimate in detail, we seem drawn to the conclusion that f_{SIG} has to be extremely small for only a few significant HGTs to have occurred.

Concluding with a related example, suppose N_{HGT} were 10^6. Then f_{SIG} would still need to be small, in this case 10^{-25}. Accordingly, unless one or more of the parameters are very different from what is assumed, Eq. (2) is consistent with significant HGT having occurred due to lightning.

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References

[1] Zimmermann U, Küppers G. Cell fusion by electromagnetic waves and its possible relevance for evolution. Naturwissenschaften 1983;70:568–9.
[2] Küppers G, Zimmermann U. Cell fusion by spark discharge and its relevance for evolutionary processes. FEBS Lett 1983;164:323–9.
[3] Demanéche S, Bertolla F, Buret F, Nalin R, Sailland A, Auriol P, et al. Laboratory-scale evidence for lightning-mediated gene transfer in soil. Appl Environ Microbiol 2001;67:3440–4.
[4] Cérémonie H, Buret F, Simonet P, Vogel TM. Isolation of lightning-competent soil bacteria. Appl Environ Microbiol 2004;70:6342–6.
[5] Cérémonie H, Buret F, Simonet P, Vogel TM. Natural electrotransformation of lightning-competent *Pseudomonas* sp. strain N3 in artificial soil microcosms. Appl Environ Microbiol 2006;72:2385–9.
[6] Kotnik T. Lightning-triggered electroporation and electrofusion as possible contributors to natural horizontal gene transfer. Phys Life Rev 2013;10(3):351–70 [in this issue].
[7] Weaver JC, Smith KC, Esser AT, Son RS, Gowrishankar T. A brief overview of electroporation pulse strength-duration space: a region where additional intracellular effects are expected. Bioelectrochemistry 2012;87:236–43.
[8] Gamerota WR, Elismé JO, Uman MA, Rakov VA. Current waveforms for lightning simulation. IEEE Trans Electromagn Compat 2012;54:880–8.
[9] Stämpfli R, Willi M. Membrane potential of a Ranvier node measured after electrical destruction of its membrane. Experientia 1957;8:297–8.
[10] Stämpfli R. Reversible electrical breakdown of the excitable membrane of a Ranvier node. An Acad Brasil Ciens 1958;30:57–63.
[11] Rubinsky B. Irreversible electroporation in medicine. Technol Cancer Res Treat 2007;6:255–60.
[12] Klenchin VA, Sukharev SI, Serov SM, Chernomordik LV, Chizmadzhev YA. Electrically induced DNA uptake by cells is a fast process involving DNA electrophoresis. Biophys J 1991;60:804–11.
[13] Sukharev SI, Klenchin VA, Serov SM, Chernomordik LV, Chizmadzhev YA. Electroporation and electrophoretic DNA transfer into cells. The effect of DNA interaction with electropores. Biophys J 1992;63:1320–7.
[14] Neumann E, Kakorin S, Tsoneva I, Nikolova B, Tomov T. Calcium-mediated DNA adsorption to yeast cells and kinetics of cell transformation by electroporation. Biophys J 1996;71:868–77.
[15] Golzio M, Teissie J, Rols MP. Direct visualization at the single-cell level of electrically mediated gene delivery. Proc Natl Acad Sci 2002;99:1292–7.
[16] Phez E, Faurie C, Golzio M, Teissie J, Rols M-P. New insights in the visualization of membrane permeabilization and DNA/membrane interaction of cells submitted to electric pulses. Biochim Biophys Acta 2005;1724:248–54.
[17] Drake FD. Discussion of space science board. West Virginia: National Academy of Sciences Conference on Extraterresterial Intelligent Life. Green Bank; 1961.
[18] Wallenhorst SG. The Drake equation re-examined. Q J R Astron Soc 1981;22:380–7.
[19] Ćirković MM. The temporal aspect of the Drake equation and SETI. arXiv:astro-ph/0306186v1.
[20] Ulmschneider PH. Intelligent life in the universe: principles and requirements behind its emergence. 2nd ed. Heidelberg: Springer; 2006.
[21] Hetesi Z, Regály Z. A new interpretation of Drake-equation. J Br Interplanet Soc 2009;59:11–4.

[22] Dominik M, Zarnecki JC. The detection of extra-terrestrial life and the consequences for science and society. Philos Trans R Soc A 2011;369:499–507.
[23] Christian HJ, Blakeslee RJ, Boccippio DJ, Boeck WL, Buechler DE, Driscoll KT, et al. Global frequency and distribution of lightning as observed from space by the optical transient detector. J Geophys Res 2003;108:4-1–4-15.
[24] Whitman WB, Coleman DC, Wiebe WJ. Prokaryotes: the unseen majority. Proc Natl Acad Sci 1998;95:6578–83.