

Supplemental Materials

Supplemental Table 1. Properties of Pluronics used in studies with PcO6 . Data for P123, P104, F108, and 25R2 are from Housely et al (2009) with surface tension measured for 0.1% solutions (v/v) at 25 °C, whereas for P68 the temperature was 23 °C (Supplemental Fig 1).

Pluronic	MW	% PEO by mass	Surface Tension	Effect on phenazine production by PcO6
P123	5750	30	34	Decrease
P104	5900	40	33	Decrease
P68	8,400	80	45	No change
F108	14,600	80	41	No change
25R2	3,100	20	43	Increase

Supplemental Table 2. Cell types to which Pluronic - F68 can serve as a cryoprotective agent under various research conditions.

Cell Types
Human erythrocytes (Glauser & Talbot, 1956)
Chinese hamster cells (Puck strain A) (Ashwood-Smith et al., 1973)
Photosynthetic picoeukaryotes (Liu et al., 2022)
Human tooth germ stem cells (Doğan et al., 2013)
<i>Oryza sativacvs</i> (Anthony et al., 1996)
<i>Lolium multiflorum</i> (Anthony et al., 1996)
Mouse BALB/c myeloma cells (González Hernández & Fischer, 2007)
Monkey African green kidney cells (González Hernández & Fischer, 2007)

Anthony, P., Jelodar, N. B., Lowe, K. C., Power, J. B., and Davey, M. R. (1996). Pluronic F-68 increases the post-thaw growth of cryopreserved plant cells. *Cryobiology*, 33(5), 508–514. <https://doi.org/10.1006/cryo.1996.0054>

Ashwood-Smith, M. J., Voss, W. A. G., and Warby, C. (1973). Cryoprotection of mammalian cells in tissue culture with pluronic polyols. *Cryobiology*, 10(6), 502–504. [https://doi.org/10.1016/S0011-2240\(73\)80004-5](https://doi.org/10.1016/S0011-2240(73)80004-5)

Doğan, A., Yalvaç, M. E., Yılmaz, A., Rizvanov, A., and Şahin, F. (2013). Effect of F68 on cryopreservation of mesenchymal stem cells derived from human tooth germ. *Applied Biochemistry and Biotechnology*, 171(7), 1819–1831. <https://doi.org/10.1007/s12010-013-0472-z>

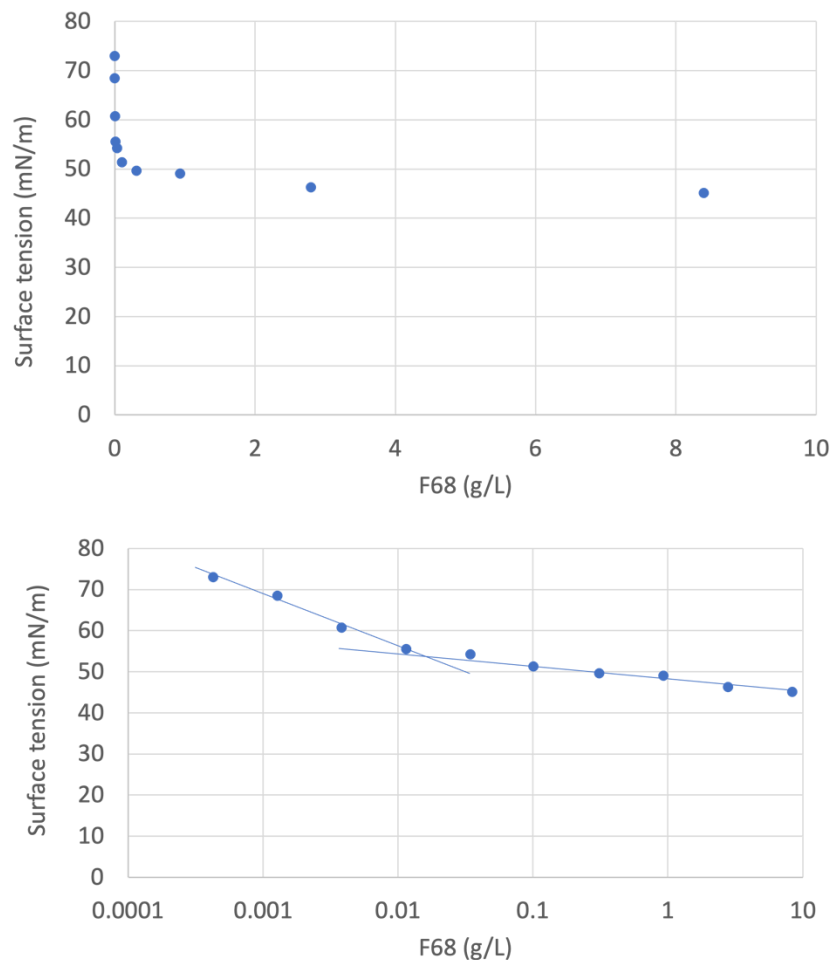
Glauser, S. C., and Talbot, T. R. (1956). Some studies on freezing and thawing human erythrocytes. *The American Journal of the Medical Sciences*, 231(1), 75–81. <https://doi.org/10.1097/00000441-195601000-00010>

González Hernández, Y., and Fischer, R. W. (2007). Serum-free culturing of mammalian cells—Adaptation to and cryopreservation in fully defined media. *ALTEX*, 24(2), 110–116.
<https://doi.org/10.14573/altex.2007.2.110>

Liu, C., Lei, J., Zhang, M., Wu, F., Ren, M., Yang, J., Wu, Q., & Shi, X. (2022). Optimization of preservation methods provides insights into photosynthetic picoeukaryotes in lakes. *Microbiology Spectrum*, 10(3), e02557-21. <https://doi.org/10.1128/spectrum.02557-21>

Supplemental Figure 1. Surfactant activity of F68.

Surfactant activity was measured at 23 °C using a wire probe tensiometer (Kibron Instruments) with defined dilutions of F68 made in double distilled water. These findings show that the surface tension values decline sharply at low concentrations before levelling off between 0.001 and 0.01 % F68.



Supplemental Figure 2

The images in Supplemental Figure 2 are controls in the studies for binding of fF68 to *PcO6* cells.

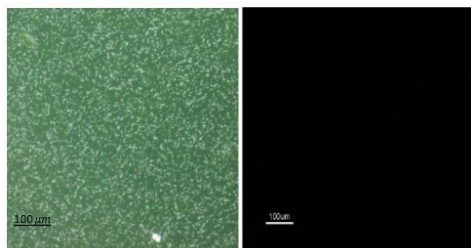
The studies were performed using the same cells as those exposed to fF68. The cells in

Supplemental Fig 1 A were without exposure to F68. In Figure set A the left-hand image is

brightfield to show the density of the *PcO6* cells which lack any native fluorescence when viewed using the FITC filter able to detect fluorescence from fluorescein, as shown in the right-hand image.

In Figure set B the left-hand image is the brightfield exposure for cells exposed to F68, without any fluorescein label for 3 h; the right-hand image in B again shows no fluorescence indicating that F68 treatment did not induce the formation of metabolites with fluorescence detected with a FITC filter set. These control studies indicate that it was the fF68 association with the *PcO6* cells that caused the exposed cells to become fluorescent when examined with the FITR filter set that detects fluorescein.

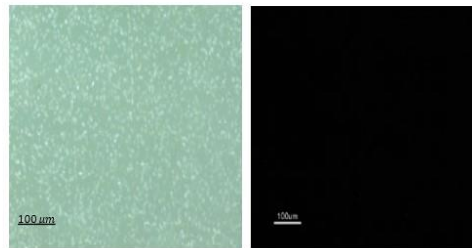
A *PcO6* cells without F68 exposure



Bright field

FITC exposure
1 sec

B *PcO6* treated with nonlabelled F68 3 h.



Bright field

FITC exposure
1 sec