EFFECTS OF PLURONIC F-68 ON 2-DEOXYGLUCOSE UPTAKE AND AMINO ACID INCORPORATION INTO CHICK EMBRYONIC FIBROBLASTS IN CULTURE

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The poloxamer surfactant, Pluronic F-68, has been used in in vitro systems to protect cells against mechanical damage (Murhammer and Goochee 1988; Handa et al 1989) and is also attracting interest as a growth-promoting supplement in such cultures (Mizrahi 1975; Bentley et al 1989). There is speculation that the effects of Pluronic involve increases in cellular nutrient uptake, but this has not been assessed in detail. We have therefore studied the effects of Pluronic F-68 cn 2-deoxyglucose (GLUC) uptake and amino acid (AA) incorporation into protein in chick embryonic fibroblast cells in culture.

Seven-day chick embryonic fibroblasts were grown at 37°C under 5% CO_2/95% air for up to 21 days in minimal essential medium supplemented with 5% horse serum, 5% foetal calf serum and antibiotics (de Pomerai and Gali 1981). Dilute cell suspensions were used for seeding cultures, which gave < 5% variation in cell numbers between replica plates. Cells were cultured with either 0.05% or 0.1% (w/v) commercial grade Pluronic F-68 (ICI/Atochem, Runcorn) or in medium alone (controls). A distinction was drawn between freshly-prepared ("fresh") Pluronic solutions and those which had been standing for 7 days or longer exposed to the air ("aged"). Cell growth was assessed by periodic measurement of cell numbers following trypsin/collagenase dissociation of replicate cultures (de Pomerai and Gali 1981). Uptake of tritiated GLUC or AA incorporation was determined over a 2h period as previously (Karim et al 1987).

During the early phases of culture growth (between 3 and 10 days), Pluronic supplementation at 0.05% (w/v), but not 0.1%, stimulated GLUC uptake by 20-30% above that in controls. This stimulatory effect was only seen when fresh Pluronic solutions were used; aged Pluronic solutions produced a 5-20% inhibition of GLUC uptake. Fresh Pluronic at both concentrations also markedly increased AA incorporation by 200-300% after 3 days of culture, but this effect was less marked after 7 days. At 10 days of culture, AA incorporation in Pluronic-supplemented cultures was slightly less than that in controls. Aged Pluronic again inhibited AA incorporation by 5-20%.

These results show that low concentrations of commercial grade Pluronic F-68 can enhance both GLUC uptake and AA incorporation into protein in cultured chick embryonic fibroblasts, provided that the solution was freshly prepared. It is tempting to speculate that these effects of Pluronic are due to interactions with the cell membrane, probably involving pore formation, as appears to be the case with microbial cells (King et al 1990). Since aged Pluronic solutions tended to inhibit uptake, we suggest that peroxide derivatives known to be formed by prolonged exposure to the air (e.g. McCoy et al 1984), may modify the cellular effects of this compound.

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