Dear Sir

We have recently measured prostaglandin (PG) production by freshly dispersed human term decidual cells in vitro.¹ To validate our protocol, we investigated the effect of a number of commercially available tissue culture media and media additives on the production of PGF₂α and PGE₂ by these cells.

Single cell suspensions of human term decidua were prepared by enzymic dispersion and Percoll density centrifugation as previously described.¹² The cells were then incubated at 2 x 10⁶ cells/ml for 2 h in various culture media with or without media supplementation. All incubations were carried out under sterile conditions at 37°C in a humidified 5% CO₂/95% air environment and in the absence of antibiotics; appropriate controls were included. PGF₂α and PGE₂ output were measured by radioimmunoassay (RIA) of the conditioned medium using antisera raised against the PGs as their methyl oximes.³ The tissue culture media investigated included RPMI 1640 containing Hepes buffer and 2 mM glutamine (Gibco-Europe, Uxbridge, Middlesex., UK), Minimal Essential Medium Eagle with Earle’s salts (Sigma Chemical Co., Poole, Dorset, UK), Dulbecco’s Modified Eagle’s medium containing 1.0 g/l glucose (DMEM; Gibco-Europe) and endotoxin-free phosphate buffered saline (PBS; Sigma); medium additives included 10% heat-inactivated normal human serum (NHS; Blood Transfusion Service, John Radcliffe Hospital, Oxford, UK), 0.25% bovine serum albumin (BSA [fraction V]; Sigma) or a serum substitute, Ultroser G, added to a final concentration of 2% as recommended (Gibco-Europe).

PG output by cells cultured in RPMI, DMEM and PBS was similar. Addition of either 10% NHS or 0.25% BSA to any of these culture media had no effect on PG output; however, the addition of 2% Ultroser G markedly decreased measured prostaglandin levels. Values are median from three separate experiments (*p < 0.05 as compared with media without 2% Ultroser G).

FIG. 1. Effect of Ultroser G on prostaglandin production by human decidual cells. Single cell suspensions of human term decidua were prepared by enzymic digestion and incubated in various culture media as described. PGF₂α (a) and PGE₂ (b) output were measured by radioimmunoassay. The presence of 2% Ultroser G (Gibco-Europe) markedly decreased measured prostaglandin levels. Values are median from three separate experiments (*p < 0.05 as compared with media without 2% Ultroser G).
Ultroser G to any of the media profoundly reduced measured levels of both PGF$_{2\alpha}$ and PGE$_2$ (Figures 1a and 1b respectively). The RIA standard curves were unaffected by the presence of 2% Ultroser G (data not shown), so it is unlikely that this medium had any affect on the performance of the assay. Moreover, 2% Ultroser G did not affect cell viability. It would seem rather that Ultroser G acts either directly or indirectly to strongly inhibit the production and/or release of PGs from term decidual cells in vitro, since attempts to stimulate PG production using the calcium ionophore, A23187 at a final concentration of 20 $\mu$M were equally frustrated (Figure 1).

Whether this phenomenon is specific to PGs and to our decidual cell model in particular remains unclear. We would suggest, however, that laboratories involved in prostanoid research should avoid the use of Ultroser G and that the possible mechanism of inhibition be further investigated.

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References

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