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Differential effect of linoleic acid on platelet aggregation

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Introduction: Linoleic acid (LA) is the most abundant omega-6 fatty acid in the diet and is generally considered to exert anti-aggregatory effects on platelets both directly (1) and when supplied as a dietary supplement (2). Whereas prolonged exposure of platelets to LA has been more commonly studied, LA may also influence platelet function by directly altering downstream eicosanoid synthesis (3). We therefore examined the acute effects of LA on platelet aggregation.

Method: Blood from healthy volunteers was collected into sodium citrate (3.2% w/v) and platelet-rich plasma (PRP) isolated by centrifugation (175*g*, 15min). In some experiments PRP was further processed into washed platelets (WP) by addition of prostacyclin (2μg/ml) and apyrase (0.02U/ml), centrifugation (2300*g*, 10min) and resuspension in modified HEPES buffer. 96-well Optimul aggregometry (4) was used to measure aggregation in response to vehicle (0.1% ascorbic acid/ethanol), LA alone (0.01 - 1mM) or LA in combination with 10μM ADP. To confirm aggregation, some WP samples were incubated with 5μM eptifibatide 5min prior the experiment. Data are presented as mean ± SEM and were analysed using two-way and one-way ANOVA with Dunnett's test, as appropriate.

Results: LA induced concentration-dependent aggregation of WP (e.g. maximal aggregation 1mM LA, 38±12%, n=9). The inclusion of eptifibatide abolished aggregation in response to all concentrations of LA (1mM LA: 6±3%, n=3, $p<0.05$). Aggregation induced by LA in PRP was greatly reduced compared to WP (1mM LA: 8±3%, n=9, $p<0.05$). When supplied in conjunction with ADP, LA caused concentration-dependent reduction in platelet aggregation in WP (vehicle, 50±9%; 1mM LA, 9±2%; n=6, $p<0.0001$) and PRP (vehicle, 89±7%; 1mM LA, 60±3%; n=7, $p<0.0001$).

Conclusions: LA stimulated concentration-dependent platelet aggregation in WP and also in PRP, although in a less pronounced manner. To determine if LA might act as a co-stimulator of platelet aggregation, platelets were co-incubated with ADP plus LA. Interestingly, LA decreased aggregation of ADP-stimulated platelets in a concentration-dependent manner. These data suggest a differential effect of LA on platelet function depending on the presence of plasma proteins and stimulus. Further research is required to elucidate the mechanisms underlying these different effects.

References:

(1) Kusznir J, Armstrong PC, Warner TD (2014). *Proceedings of the British Pharmacological Society* at http://www.pA2online.org/abstracts/Vol12Issue3abst174P.pdf
(2) Challen AD, Branch WJ and Cummings JH (1983). *Clinical Nutrition* **37**: 197–208.
(3) Srivastava KC (1985). *Prostaglandins Leukot Med.* **17**: 319-27.
(4) Chan MV, Armstrong PC, Papalia F, Kirkby NS and Warner TD (2011). *Platelets* **22**: 485-94.