Mechanisms of Platelet Inhibition by the Selective Serotonin Uptake Inhibitor Citalopram

Background: Selective serotonin reuptake inhibitors (SSRIs) are commonly prescribed anti-depressants that block serotonin uptake by the serotonin reuptake transporter (SERT). Since platelets also express SERT, SSRIs can affect platelet function and may also therefore confer benefit against myocardial infarction (1). However, the cardiovascular benefits and risks of SSRIs are poorly characterised and useful in vitro platelet data is limited. The SSRI citalopram is a racemic (RS) mixture and (S)-citalopram has been reported to be 27 times more potent than (R)-citalopram for SERT blockade (2). Citalopram has previously been reported to inhibit collagen-induced platelet activation in vitro(3), although whether this effect is mediated directly through blocking SERT or by some alternative mechanism is unclear.

Aim: To determine if SERT blockade is the mechanism by which citalopram inhibits platelet function.

Methods: Citrated fresh blood was collected from healthy human volunteers and washed platelets prepared (2 x 10^8 ml^-1), before incubation with either racemic citalopram or its isomers (1 nM – 200 μM). Collagen-induced platelet aggregation was measured using two optical AggRAM aggrego meters. Both rate and extent of aggregation were quantified from individual aggregation traces. SERT activity was measured by adding exogenous serotonin (1 μM) to 5 ml washed platelets and determining the concentration of serotonin in the supernatant of aliquots drawn at 5 min time intervals for up to 1 hr. Serotonin concentrations were subsequently quantified using high pressure liquid chromatography. A four parameter logistic model was used to fit concentration-response data using NONMEM 7.3.0® for serotonin uptake data or MS Excel®.

Results: The rate of aggregation induced by a maximal concentration of collagen in the absence of citalopram was 189.1 ± 10.18%.min^-1. Citalopram and its isomers inhibited this maximal rate with similar pIC50 values: (RS) 4.00 ±0.06; (S) 3.97 ± 0.10; (R) 4.06 ± 0.08. This represents an S:R potency ratio of 0.91. The concentration-inhibition relationship was steep (Hill coefficient:(RS) 5.1 ±1.3; (S) 2.6 ± 0.8; (R) 7.6 ± 3.7). Following addition to platelets, serotonin concentrations fell in an exponential manner indicating a first order mechanism, and rate constants for uptake (k_u) were calculated. In the absence of citalopram, k_u = 4.6 ± 0.2hr^-1 (t_1/2 ~ 9 min). Uptake was abolished by each citalopram isomer at concentrations exceeding 100 nM and the concentration-inhibition curves had Hill coefficients that were not significantly different from 1 (P = 0.099). The pIC50 values were: (RS) 8.33 ± 0.03; (S) 8.67 ± 0.04; (R) 7.38 ± 0.04. This represents an S:R potency ratio of 19.5 which is comparable to previous reports for SERT inhibition (2) but contrasts markedly with the aggregation inhibitory potencies.

Conclusions: Serotonin uptake into platelets was completely blocked by each citalopram isomer at concentrations that had no effect on platelet aggregation (1-10 μM). This observation and the different S:R potency ratios strongly suggest that SERT blockade is not the primary mechanism by which citalopram inhibits platelet aggregation. We conclude that inhibition of platelet aggregation by citalopram is not mediated via SERT blockade and alternative mechanisms of action are more likely. This conclusion presents two additional questions: by what mechanism does citalopram inhibit platelets, and what effect does SERT inhibition have on platelet and cardiovascular function, if any?

References:

