Endocytosis of plasma-derived factor V by megakaryocytes to yield the unique platelet-derived cofactor molecule is mediated by residues within the factor V light chain

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Platelet- and plasma-derived factor Va serve as essential cofactors for thrombin generation at sites of vascular injury. However, the platelet-derived molecule is physically and functionally distinct from its plasma counterpart, and forms the more hemostatically-relevant pool. Despite these differences, this unique factor Va pool originates *via* endocytosis and modification of the plasma-derived procofactor, factor V, by platelet precursors, megakaryocytes. Studies from our laboratory have shown that factor V endocytosis requires expression of two receptors: an unidentified, factor V-specific receptor and LDL receptor related protein-1 (LRP-1). These combined observations represent a unique role for LRP-1 in endocytosis of a protein that is modified functionally and not destined for lysosomal degradation. The goal of this study is to define the factor V residues involved in its interactions with the two receptor system. ^{125}I-factor V endocytosis assays, using various factor V molecules, as well as isolated factor V domains, indicated that factor V endocytosis is inhibited by the factor V light chain. Consistent with this observation, endocytosis was also inhibited by several anti-factor V light chain antibodies. Additional experiments indicated that one of the inhibitory antibodies, anti-FV#2, is against an epitope near or including Arg^{1765}/Leu^{1766}, the factor Xa cleavage site in the factor V light chain. As this region of factor V has sequence homology common to other LRP-1 ligands, it is anticipated that it is involved in LRP-1 binding. However, additional studies are necessary to determine the receptor with which this region of factor V interacts. Furthermore, synthetic factor V peptides spanning the factor Xa cleavage site will be tested. As platelet-derived factor Va represents the more physiologically-relevant pool, understanding the mechanisms regulating its formation are of interest. Further studies will provide a more complete understanding of this unique receptor system, which could potentially serve as a target for therapeutic intervention.