Complex Interactions of excitatory and inhibitory stimuli in the vascular endothelium

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The endothelium is the single, innermost, layer of cells in blood vessels and it controls almost all cardiovascular functions. The endothelium regulates immune responses, blood coagulation, angiogenesis, vessel repair, and vascular permeability. To regulate function, the endothelium continuously monitors multiple chemicals that are circulating in the blood or derived from nearby cells (e.g. endocrine, paracrine, autocrine or neurotransmitter signals). These chemicals each provide cues to physiological status. The chemicals are detected and processed selectively by the endothelium to direct resources that adjust physiological function. Some chemicals provide conflicting signals to the endothelium such as those which promote or inhibit vascular tone. How conflicting signals are detected and managed by the endothelium is not understood. The present study was undertaken to determine how the endothelium selectively processes conflicting information. To monitor the behaviour of the endothelium, changes in endothelial cytosolic Ca2+ concentration were measured in ~200 endothelial cells in small mesenteric arteries using the indicator Cal-520 and high-resolution imaging. Changes in cytosolic Ca2+ concentration underlies virtually all endothelial functions and so provides a useful measure of endothelial activity. The vasodilator substance acetylcholine evoked Ca2+ increases that initiated in small clusters of cells. As the concentration of acetylcholine increased, the amplitude of the Ca2+ rise and the number of activated cells increased. To examine the interaction of excitatory and inhibitory inputs, acetylcholine (as an excitatory input) at an EC75 (15nM) was used. In the first series of experiments the vasoconstrictor alpha-adreneric agonist phenylephrine was used. Phenylephrine (10 μM) did not itself evoke any detectable Ca2+ signals in the endothelium. However, phenylephrine inhibited the Ca2+ rise evoked by acetylcholine (15 nM). Phenylephrine inhibited the amplitude of the response in each activated cell and the number of cells activated. By inhibiting endothelial activity, phenylephrine will increase the contractile response operating on the smooth muscle cells. The vasoconstrictor agonist angiotensin (10 μM) did not evoke Ca2+ increases when applied by itself. However, angiotensin increased the amplitude of the Ca2+ response to acetylcholine in each activated cell. The number of cells activated did not change. This effect of angiotensin on the endothelium may serve to limit a vasoconstrictor response. Together these data reveal complex interaction occur between excitatory and inhibitory stimuli acting on the endothelium.

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