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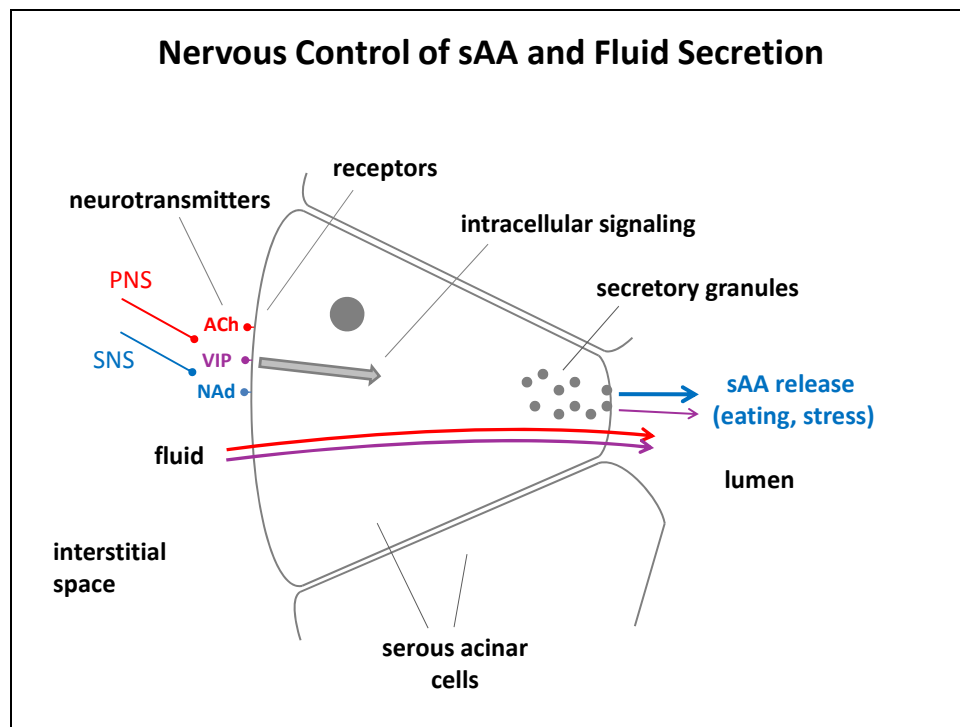
## General Information on Salivary Proteins

Numerous proteins and peptides are secreted by the saliva glands into the oral cavity to serve a variety of functions. During periods between meals, glycoproteins known as mucins play a key role in moistening, lubricating, and protecting the mouth. Various other proteins that serve protective or anti-microbial roles in the mouth, including salivary alpha-amylase (sAA), are also secreted in resting saliva. (Veerman et al., 1996; Nieuw Amerongen et al., 2007; Proctor & Carpenter, 1998). Once the salivary reflex is stimulated by taste or motions of the jaw, however, large amounts of mucins are needed to lubricate the food for chewing and swallowing. Secretion of sAA—whose main function is as an enzyme that breaks down starches—also increases substantially. Due to the need for rapid release of large quantities of these proteins, they are synthesized in the hours between meals and stored in secretory granules within the cells of the salivary glands. These granules congregate near the cell membrane that faces the interior lumen of the gland, where they can quickly release the stored proteins into the lumen by a process known as exocytosis. (Proctor & Carpenter, 2007; Proctor & Carpenter, 1998; Gorr et al., 2005; Turner & Sugiya, 2002)

Saliva also contains some proteins that are not produced in the salivary glands. Serum proteins such as albumin are generally too large to move through salivary cell membranes; when present in whole saliva they enter largely through gingival crevicular fluid or through injuries to mucosal tissues. (Nieuw Amerongen et al., 2007; Vining et al., 1983) Whole saliva also contains enzymes and other products from oral bacteria. (Kaufman & Lamster, 2002). Some other proteins (e.g., IgA, insulin) are present in glandular saliva due to active transport mechanisms. The transport of IgA into saliva has been studied and has been shown to be affected by autonomic activity. (Brandtzaeg, 2007; Groschl, 2008; Proctor & Carpenter, 2001)

Most of the proteins and peptides present in saliva are synthesized in the acinar (or occasionally ductal) salivary cells. (Proctor & Carpenter, 1998) The secretion of these proteins in saliva is controlled in a cooperative fashion by the sympathetic and parasympathetic innervation of the salivary glands. Neurotransmitters released from the nerves bind to receptors on the cells, initiating intracellular signaling pathways that lead to release of proteins from the storage granules. Release of mucins from the mucous acinar cells of minor and sublingual glands is largely stimulated by release of acetylcholine (ACh) and the neuropeptide VIP from parasympathetic

nerves. In contrast, release of various proteins, including sAA, from serous acinar cells in the submandibular and parotid glands is mainly controlled by release of noradrenaline from sympathetic nerves. Additionally, however, parasympathetic stimulation, acting through neuropeptide transmitters such as VIP, can also contribute to significant secretion of alpha-amylase. Release of ACh from parasympathetic nerves also triggers intracellular signaling that increases fluid flow through the cells. Fluid flow is further augmented by the release of VIP from parasympathetic nerves. (Proctor & Carpenter, 2007; Culp & Richardson, 1996; Ekström, 1999; Ambudkar, 2011) Since increased fluid secretion often accompanies protein secretion, saliva flow needs to be considered when measuring concentrations of proteins such as sAA in saliva. (Bosch et al., 2011)



In addition to the changes in saliva flow and protein secretion that are tied to reflex stimulation associated with eating, saliva flow and protein levels are also affected by activity in higher centers of the brain. The association of reduced saliva flow with anxiety has long been recognized, and studies in the 1990s demonstrated that sAA levels in saliva respond to both physical and mental stressors. (Proctor & Carpenter, 2007; Chatterton et al., 1996; Chatterton et al., 1997) sAA has since become the best known of the salivary proteins to behavioral researchers because its release into saliva has promoted as a marker of SNS activity. (Rohleder et al., 2004,

Granger et al., 2007, Nater & Rohleder, 2009) However, recent discussions have re-emphasized evidence from earlier studies that demonstrated that sAA release is also partially under PNS control. (Bosch et al., 2011, Nater & Rohleder, 2009)

Salivary proteins other than sAA are also secreted in response to autonomic signals, and it is possible that some of these could be found to have uses as autonomic biomarkers. For example, two revealing studies by Bosch et al. (2001; 2003) have illustrated how saliva flow and secretion of various salivary proteins (SIgA, mucins, cystatin S, and lactoferrin) are differently affected by sympathetic and parasympathetic responses to different types of stressors. Two proteins that have been studied in blood and that are also released into saliva are chromogranin A (CgA) and nerve growth factor (NGF); it is thought that these may have potential as salivary markers of autonomic nervous activity. (See links to CgA and NGF in this issue for more information.)

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