Somatostatin system: molecular mechanisms regulating anterior pituitary hormones

Tamar Eigler and **Anat Ben-Shlomo**

Division of Endocrinology, Diabetes and Metabolism, Department of Medicine, Pituitary Center, Cedars Sinai Medical Center, Davis Building, Room 3066, 8700 Beverly Boulevard, Los Angeles, California 90048, USA

Correspondence should be addressed to A Ben-Shlomo
Email
benshlomoa@cshs.org

Abstract

The somatostatin (SRIF) system, which includes the SRIF ligand and receptors, regulates anterior pituitary gland function, mainly inhibiting hormone secretion and to some extent pituitary tumor cell growth. SRIF-14 via its cognate G-protein-coupled receptors (subtypes 1–5) activates multiple cellular signaling pathways including adenylate cyclase/cAMP, MAPK, ion channel-dependent pathways, and others. In addition, recent data have suggested SRIF-independent constitutive SRIF receptor activity responsible for GH and ACTH inhibition *in vitro*. This review summarizes current knowledge on ligand-dependent and independent SRIF receptor molecular and functional effects on hormone-secreting cells in the anterior pituitary gland.

Key Words
- somatostatin
- receptors
- pituitary
- hormone secretion

Journal of Molecular Endocrinology
(2014) **53**, R1–R19

Journal of Molecular Endocrinology

Introduction

The anterior pituitary gland is subjected to the stimulatory and inhibitory effects of multiple regulators. Somatostatin (SRIF) and its cognate receptors (sst1–sst5) exhibit a dominant inhibitory role in pituitary gland regulation. Hypothalamic SRIF was isolated from the hypothalamus (Burgus *et al*. 1973) and subsequently demonstrated to be secreted throughout the brain and from multiple peripheral organs, affecting multiple tissues (Patel 1999).

The pituitary gland is positioned outside the blood–brain barrier, and is composed of two entities that merge during embryonic development, the anterior and intermediate lobes that ascend from the oral ectoderm and the posterior lobe that descends from the hypothalamus (Drouin 2011). The anterior pituitary harbors hormone-secreting epithelial-origin cell types, including those expressing prolactin (PRL) and growth hormone (GH) that compose most of the gland, centrally located adrenocorticotropin (ACTH)-secreting and thyrotropin (TSH)-secreting cells, and laterally scattered gonadotropin (follicle-stimulating hormone (FSH) and luteinizing hormone (LH)) cells. The intermediate lobe contains cells secreting α-melanotropin; however, this lobe degenerates in humans. The posterior lobe harbors axons descending from neurons located in the hypothalamic nuclei and release vasopressin (antidiuretic hormone) and oxytocin (Bichet 2011). As various SRIF receptor expression levels and subtype profiles were observed on all pituitary cell types (Ben-Shlomo & Melmed 2010), a range of SRIF system effects are exhibited in the different cell types.

Cortistatin (CST), a ligand with SSTR binding affinity similar to that of SRIF, is expressed in the cerebral cortex and hippocampus, but not in the hypothalamus (Spier & de Lecea 2000); hence, it is not a major endocrine regulator of pituitary signaling and function.

SRIF receptors (SSTRs) exhibit *in vitro* constitutive activity, independently of SRIF or CST presence, and

http://jme.endocrinology-journals.org
DOI: 10.1530/JME-14-0034

Published by Bioscientifica Ltd.

regulate GH and ACTH production (Ben-Shlomo *et al.* 2009, 2013). The SRIF ligand and the five SRIF receptor subtypes (sst1–sst5) regulate pituitary function at two levels, via ligand exposure and potentially via selective receptors, independently of the ligand.

Somatostatin

Somatostatinergic neuronal cell bodies lie within the anterior periventricular nucleus and comprise 80% of hypothalamic SRIF immunoreactivity. The remaining hypothalamic SRIF-producing neuronal bodies lie within the paraventricular, arcuate, and ventromedial nuclei. Retrograde-tracing functional topography of hypothalamic SRIF neurons in the male rat demonstrated that SRIF neurons regulating the pituitary are confined within the periventricular and paraventricular nuclei, but not in the arcuate nucleus (Kawano & Daikoku 1988). These neurons send axonal projections to the median eminence at the base of the hypothalamus (Fig. 1). Ultrastructural morphometric analysis of SRIF-like immunoreactive neurons indicated that more than half of all terminals in the median eminence exhibit SRIF-containing vesicles with estimated 0.7 mM concentration per vesicle (Foster & Johansson 1985). Hypothalamic SRIF neuron axons descend from the median eminence toward the pituitary stalk and terminate at the pituitary portal blood vessel system, releasing SRIF into the blood reaching the anterior pituitary cells (Patel 1999) or travel through the neural pituitary stalk into the posterior pituitary (Patel & Srikant 1986; Fig. 2).

SRIF is cleaved from a common SRIF prohormone into several cyclic tetradecapeptide products by prohormone

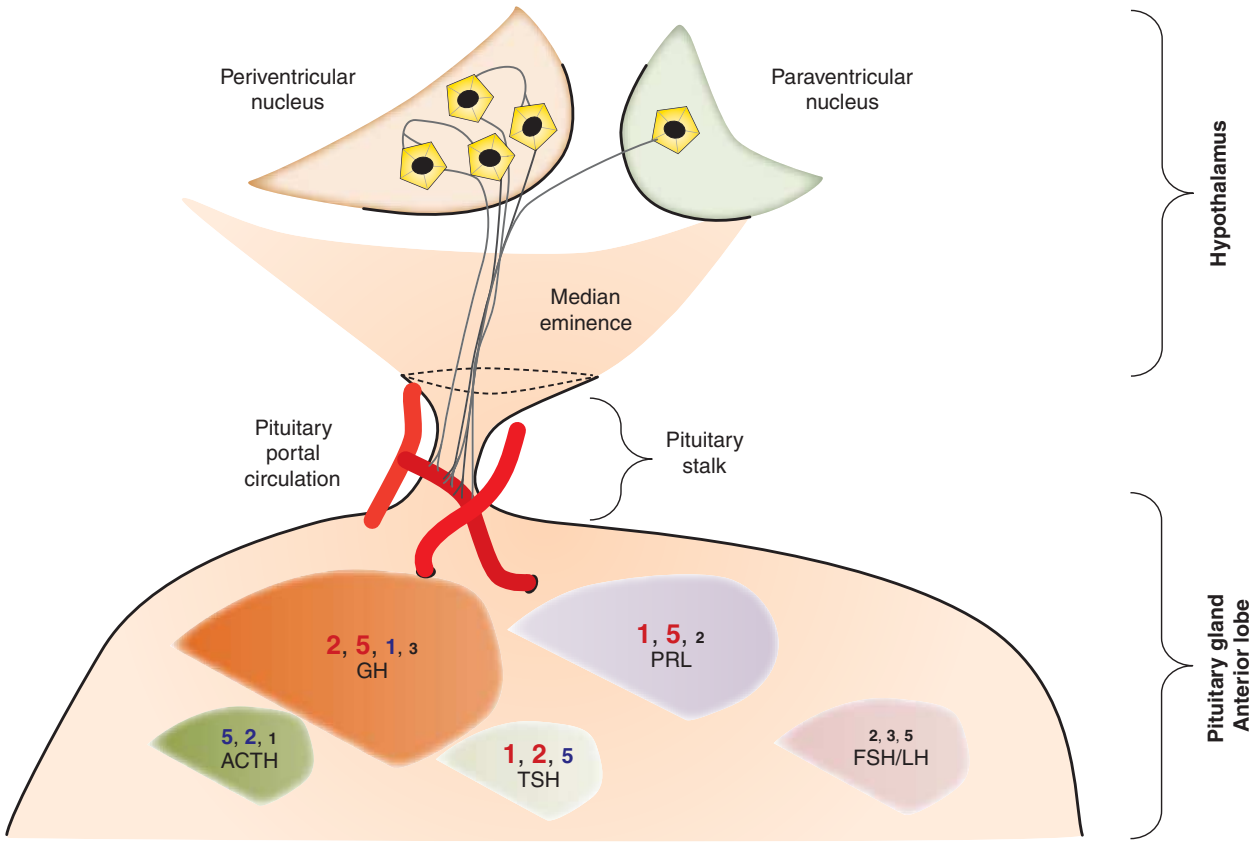

Figure 1

Somatostatin system: hypothalamus–anterior pituitary lobe axis. Somatostatinergic neuron bodies located at the hypothalamic periventricular (80%) and paraventricular (20%) nuclei travel through the median eminence and pituitary stalk and secrete SRIF into the pituitary portal circulation, reaching the cells of the anterior pituitary gland. Receptor subtype distribution is represented by subtype numbers (1–5) inside the specific cell type and is based on receptor profile in cell-respective human tumors, i.e. GH-, ACTH-, PRL, TSH-secreting adenomas and non-functioning pituitary tumors that also include gonadotroph FSH/LH secreting tumors. Large size red letter indicates that ~90% of selective tumor type are positive for that receptor subtype, medium size blue letter indicate that ~70% of selective tumor types are positive for that receptor subtype, small size black letters that ~50% of selective tumor type are positive for that receptor subtype. Lower percentages are not presented. Receptor profile data is based on Ben-Shlomo & Melmed (2010).

http://jme.endocrinology-journals.org
DOI: 10.1530/JME-14-0034

Published by Bioscientifica Ltd.

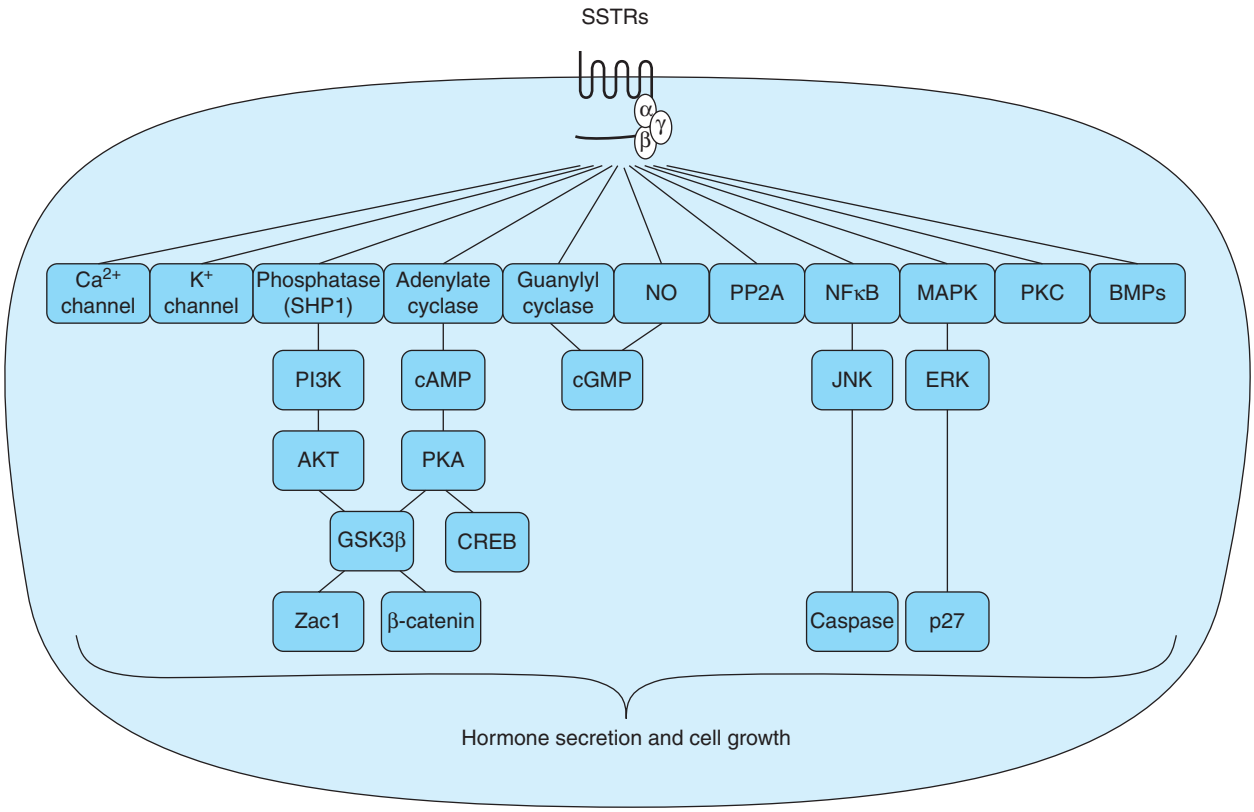

Figure 2

Selected SRIF-dependent pituitary signaling pathways. SRIF and SRIF analogs activate multiple molecular signaling pathways depicted here, which control pituitary hormone secretion as well as cell growth. These include Ca^{2+} and K^{+} channels, phosphatases such as SHP1 and PP2A, cyclic nucleotide synthases such as guanylyl and adenylyl cyclase, nitric oxide, NFκB, MAPK/ERK, PKC, as well as BMPs.

convertases (Galanopoulou *et al*. 1995); however, SRIF-14, which contains 14 amino acids, is the predominant form of SRIF in the brain, including the hypothalamus (Acunzo *et al*. 2008), and therefore the predominant pituitary regulator.

Multiple factors regulate hypothalamic SRIF-14 production and secretion. Table 1 lists factors demonstrated to have a direct effect on hypothalamic SRIF production and/or secretion. Of note, most studies utilized either *ex vivo* hypothalamic slices or hypothalamic primary cell cultures, attempting to isolate the effects of the studied molecule on SRIF.

SRIF half-life is short (~2 min) as it is rapidly internalized and inactivated by peptidases inside the cell after internalization (Roosterman *et al*. 2008) and in the circulation (Werle & Bernkop-Schnurch 2006). To overcome this limitation for clinical use, analogs such as octreotide (Bauer *et al*. 1982), lanreotide (Sassolas *et al*. 1989), and pasireotide (Bruns *et al*. 2002) were synthesized as stable SRIF agonists.

SRIF regulates pituitary function through the G-protein-coupled receptors (GPCRs): SRIF receptor subtype 1 (sst1), sst2, sst3, and sst5. The expression of sst4 in the normal adult pituitary gland remains unclear. Although sst2 is alternatively spliced to sst2a and sst2b, only the sst2a isoform is expressed in the human pituitary tumors (Panetta & Patel 1995). The five human SSTR genes are located on five different chromosomes and encode receptor protein of size ranging from 356 to 391 amino acid residues with 39–57% sequence identity among the receptors (homology derives mostly from the transmembranal domain; Patel 1999). Multiple factors that regulate *SSTR* expression levels (Ben-Shlomo & Melmed 2010) are provided in Table 2.

SRIF-14 exhibits high binding affinity (few hundreds pM in membrane extracts from cell transfectants *in vitro*) to all receptor subtypes (Patel 1999). Upon ligand binding, the receptors bind the $G_{\alpha i/o}$ subunit of the $G_{\alpha\beta\gamma}$ tetramer, releasing $G_{\beta\gamma}$, initiating multiple cascades of signaling pathways. Most studies on SRIF-14 regulation of pituitary

http://jme.endocrinology-journals.org
DOI: 10.1530/JME-14-0034

Published by Bioscientifica Ltd.

Table 1 Molecules regulating hypothalamic SRIF production

Effect	Molecule	Method utilized[a]	References
Stimulation	Acetylcholine	Rat fetal hypothalamic primary cultures (d18)	Peterfreund & Vale (1983)
	Dopamine	Male rat hypothalamic segments	Negro-Vilar *et al*. (1978), Maeda & Frohman (1980) and Lengyel *et al*. (1985)
	Neurotensin	Rat hypothalamic segments	Sheppard *et al*. (1979), Maeda & Frohman (1980) and Shimatsu *et al*. (1982)
	Melatonin	Rat hypothalamic segments	Richardson *et al*. (1981)
	Glucagon	Perfused hypothalamic halves of male rats	Shimatsu *et al*. (1982)
	Growth hormone	Rat hypothalamic segments	Sheppard *et al*. (1978)
	IGF1	Rat hypothalamic segments	Berelowitz *et al*. (1981)
	Sex steroids[b]	*In vivo* and *in vitro* approaches. SRIF mRNA, protein, or hypothalamic neuron number	Werner *et al*. (1988), Zorrilla *et al*. (1990), Senaris *et al*. (1992), Simonian *et al*. (1998), Pillon *et al*. (2004) and Zhang *et al*. (2009)
	Thyroid hormones	Rat hypothalamic segments	Berelowitz *et al*. (1980)
	Insulin	Rat hypothalamic segments	Berelowitz *et al*. (1982)
	GHRH	Cultured fetal rat hypothalamic cells	Iwasaki *et al*. (1987) and Richardson *et al*. (1988)
	CRH	Cultured fetal rat hypothalamic cells	Iwasaki *et al*. (1987)
	TRH	Cultured fetal rat hypothalamic cells	Iwasaki *et al*. (1987)
	NPY	Rat hypothalamic segments	Korbonits *et al*. (1999)
	Bombesin	SRIF in the hypophysial portal blood	Abe *et al*. (1981)
	Norepinephrine	Dispersed adult male rat hypothalamic cells	Negro-Vilar *et al*. (1978) and Richardson & Twente (1990)
	Substance P	Rat hypothalamic segments	Sheppard *et al*. (1979)
	Cytokines: IL1 and IL2	Dispersed fetal rat diencephalic cells; mediobasal hypothalamus section	Scarborough *et al*. (1989), Honegger *et al*. (1991) and Karanth *et al*. (1993)
Inhibition	γ-aminobutyric acid (GABA)	Rat fetal hypothalamic primary cultures (d18)	Peterfreund & Vale (1983)
	Serotonin	Rat fetal hypothalamic primary cultures (d18)	Richardson *et al*. (1981) and Peterfreund & Vale (1983)
	Acetylcholine	Male rat hypothalamic segments	Richardson *et al*. (1980)
	Vasoactive intestinal polypeptide (VIP)	Perfused hypothalamic halves of male rats	Shimatsu *et al*. (1982)
	Leptin	Fetal rat neurons in monolayer culture	Quintela *et al*. (1997*a*)
	Somatostatin	Rat hypothalamic periventricular nucleus fragments	Aguila (1998)
	Opioids	Rat hypothalamic fragments	Lengyel *et al*. (1985)
	Glucose	Rat hypothalamic segments	Berelowitz *et al*. (1982)
	Cytokines: TGFβ	Primary monolayer cultures of hypothalamic cells	Quintela *et al*. (1997*b*)
Dual	Glucocorticoids	*In vivo* injection induced while *in vitro* hypothalamic segment perfusion decreased SRIF secretion	Estupina *et al*. (1997)

[a]All experiments were carrried out in male rats.
[b]Some report decreased somatostatin level following sex steroid treatment (Fernandez *et al*. 1992, Hassan *et al*. 2001). This discrepance may be due to the experimental approach including whether treatment was conducted *in vivo* or *in vitro*, treatment duration and dose, sex of the animal model and species.

function have focused on sst2 and sst5; however, the adult pituitary gland expresses sst1 and sst3 that are yet to be explored in this context. Although SRIF signaling pathways in non-pituitary cells have been extensively investigated, with more than 20 such intracellular pathways described (Cervia & Bagnoli 2007), pituitary SRIF-mediated molecular signaling pathways has been mostly limited to ion channel regulation, adenylate cyclase/cAMP/PKA regulated pathways, and protein phosphatase activation (Ben-Shlomo & Melmed 2010).

SRIF-dependent pituitary molecular signaling pathways

SRIF-14 signaling in the anterior pituitary gland primarily mediates the regulation of hormone secretion, yet also plays a role in regulation of cell growth (Fig. 1).

Ion channel regulation

The dominant function of SRIF-dependent pituitary signaling is the inhibition of stimulated hormone secretion.

http://jme.endocrinology-journals.org
DOI: 10.1530/JME-14-0034

Published by Bioscientifica Ltd.

Table 2 Factors regulating pituitary cell SRIF receptor expression[a]. Table adapted and updated from *Trends in Endocrinology & Metabolism*, 21, A Ben-Shlomo & S Melmed, Pituitary somatostatin receptor signaling, 123–133, copyright (2010), with permission from Elsevier

Treatment	sstr-1	sstr-2	sstr-3	sstr-4	sstr-5	References
SRIF 14 (>24 h)	+	+	+	+	+	Berelowitz *et al*. (1995) and Luque *et al*. (2004)
High dose SRIF (4 h)	+	+	0	0	+	Cordoba-Chacon *et al*. (2012)
Low dose SRIF (4 h)	+	0	0	0	−	Cordoba-Chacon *et al*. (2012)
Forskolin	+	+	NA	NA	0/−	Patel *et al*. (1993), Luque *et al*. (2004) and Cordoba-Chacon *et al*. (2012)
PKC activator (TPA)	+	0	0	0	0	Cordoba-Chacon *et al*. (2012)
GHRH	0/+	0/+	NA	NA	0/−	Luque *et al*. (2004), Park *et al*. (2004) and Cordoba-Chacon *et al*. (2012)
Ghrelin	0/+	0/−	NA	NA	0/−	Luque *et al*. (2004), Yan *et al*. (2004) and Cordoba-Chacon *et al*. (2012)
17β-estradiol	−/+	+	+	NA	+/−	Xu *et al*. (1995), Djordjijevic *et al*. (1998), Kimura *et al*. (1998), Canosa *et al*. (2003) and Cardenas *et al*. (2003)
Testosterone	0/+	0/+	+	NA	0	Xu *et al*. (1995)
Thyroxine	+	NA	NA	NA	+	James *et al*. (1997)
Glucocorticoids (2 h)	+	+	0	NA	NA	Xu *et al*. (1995)
Glucocorticoids (24–48 h)	−	−	− then +	NA	0	Xu *et al*. (1995), Petersenn *et al*. (1999) and van der Hoek *et al*. (2005)
Progesterone	+	0	−	NA	NA	Xu *et al*. (1995)
Food deprivation	−	−	−	0	0	Berelowitz *et al*. (1995)
Diabetes mellitus	−	−	−	0	−	Berelowitz *et al*. (1995)
TGFβ	NA	+	NA	NA	NA	Puente *et al*. (2001)

+, Upregulation of receptor expression; −, downregulation of receptor expression; 0, no change in receptor expression; NA, not assessed.
[a]This table incorporates results from different species (pig, rat, and fish, baboon), genders, and assay techniques (*in vitro* and *in vivo*, primary cultures and cell-lines, mRNA transcripts, and promoter activation measurements); therefore an integrated interpretation is difficult.

Hypothalamic hormones (Bjoro *et al*. 1987, Spada *et al*. 1990, Bonnefont *et al*. 2000, Liu *et al*. 2006, Tsaneva-Atanasova *et al*. 2007) signal to release anterior pituitary hormone secretion by increasing intracellular Ca^{2+} levels, resulting in the exocytosis of hormone-containing vesicles. Most information related to SRIF-mediated ion channel regulation was accrued through the investigation of mechanisms for SRIF-dependent inhibition of GH secretion. Although the definitive role of individual ion channels in regulation of hormone secretion are not fully understood, advances have been made through the use of electro-physical methods in addition to molecular biological approaches. Na+, Ca^{2+}, and K^+ channels have been isolated in somatotroph cell membranes and described to contribute to hormone secretion.

GH secretion is activated upon GH-releasing hormone (GHRH) binding and activation of somatotroph cell surface receptors. GHRH signaling causes membrane depolarization and an action potential burst in response to the opening of tetrodotoxin-insensitive Na^+ channels. In turn, increased Ca^{2+} transient frequency and intracellular Ca^{2+} concentration lead to amplified exocytosis of GH-containing granules (Tsaneva-Atanasova *et al*. 2007). In contrast, SRIF antagonizes the effect of GHRH through membrane hyperpolarization by opening K^+ channels leading to depletion of intracellular Ca^{2+} concentration, effectively inhibiting GH exocytosis (Kraicer & Spence 1981, Draznin *et al*. 1988, White *et al*. 1991, Tsaneva-Atanasova *et al*. 2007). SRIF signaling through sst2 and sst4 activates K^+ influx through both inwardly rectifying channel conductance and delayed rectifying K^+ channels in GH_3 cells (Yang *et al*. 2005, 2007, Yang & Chen 2007). SRIF targets the large-conductance, calcium- and voltage-activated K^+ channels (BK channel) in GH_4C_1 cells (White *et al*. 1993). Ultimately, these effects result in membrane hyperpolarization and closure of L- and N-type voltage sensitive calcium channels (Petrucci *et al*. 2000, Cervia *et al*. 2002*a*, Tsaneva-Atanasova *et al*. 2007, Yang *et al*. 2007). SRIF-reduced T-type current occurs primarily in rat somatotroph cultures (Chen *et al*. 1990, Yang *et al*. 2007). In somatotroph cells, SRIF regulation of K^+ currents are mediated by $G\alpha_{i3}$ (Chen 1997), while Ca^{2+} currents are mediated by $G\alpha_{o2}$ (Chen 1997, Degtiar *et al*. 1997), β1, β3 (Kleuss *et al*. 1992), and γ3 (Kleuss *et al*. 1993). In human GH-secreting tumor cultures, sst5-specific signaling is dependent on $G\alpha_{o1}$ (Peverelli *et al*. 2013).

SRIF-mediated inhibition of corticotropin-releasing hormone (CRH)-stimulated Ca^{2+} levels in human corticotropin-secreting pituitary adenomas is

http://jme.endocrinology-journals.org
DOI: 10.1530/JME-14-0034

Published by Bioscientifica Ltd.

effectively blocked by pretreatment with pertussis toxin (PTX), indicating that the effect is $G\alpha_{i/o}$ dependent (Spada *et al*. 1990). SRIF-induced K^+ influx is regulated by $G\alpha_{i3}$ in AtT-20 corticotroph cells (Takano *et al*. 1997). Although it remains to be determined whether SRIF regulates pituitary Ca^{2+} levels independently of K^+ channels, GH release correlates with both frequency and amplitude of calcium oscillations, while calcium channel blockers and SRIF acutely suppress Ca^{2+} extrusion (Holl *et al*. 1988). SRIF treatment resulted in Ca^{2+}-dependent redistribution of cytoplasmic microfilaments, without affecting intracellular somatotroph GH content (Shimada *et al*. 1990), in addition to reduced association of exocytosis-associated RAB3B and SNARE proteins (Matsuno *et al*. 2003). Finally, it has been recently shown that SRIF inhibited *CaMKiiβ* expression and protein levels, and knockdown of *CaMKiiβ* decreases Ca^{2+} levels in GC cells and suppressed secretion, suggesting that *CaMKiiβ* may mediate SRIF regulation of Ca^{2+} (Cervia 2011). In summary, although the role of SRIF-dependent regulation of ion channels in pituitary cell growth is unknown, SRIF-dependent regulation K^+-derived membrane hyperpolarization and the reduction of Ca^{2+} influx and concentration mediate the acute regulation of the exocytosis of hormone-containing vesicles.

Adenylate cyclase/cAMP/PKA signalling

SRIF inhibits pituitary adenylate cyclase/cAMP/PKA signaling, thereby inhibiting pituitary hormone synthesis and cell growth. SRIF inhibits cAMP production and ACTH secretion induced by CRH, forskolin, isoproterenol, vasoactive intestinal polypeptide (VIP), and cholera toxin in AtT-20 cells (Heisler *et al*. 1982). Similarly, SRIF inhibits cAMP and GH production induced by GHRH stimulation in primary pituitary cells (Bilezikjian & Vale 1983). SRIF inhibits forskolin-induced cAMP/PKA signaling pathway in rat somatotropic cells (Tentler *et al*. 1997). sst1, sst2, sst3, and sst5 all mediate SRIF inhibition of adenylate cyclase in pituitary cells (Tentler *et al*. 1997, Cervia *et al*. 2003, Ben-Shlomo *et al*. 2005). SRIF inhibited forskolin-induced cAMP production, PKA activation, CREB phosphorylation, and transcription potency, while overexpression of the PKA catalytic subunit suppressed SRIF action in sst2 stable transfectant GH_4 cells (Tentler *et al*. 1997).

The ability of SRIF to inhibit adenylate cyclase is $G_{\alpha i/o}$-dependent (Koch *et al*. 1985, Tallent & Reisine 1992, Liu *et al*. 1994, Morishita *et al*. 2003). In GH_4C_1 cells, SRIF-action is mediated specifically via $G\alpha_{i2}$ (Liu *et al*. 1994), as PTX suppresses SRIF-mediated inhibition of VIP-induced cAMP (Koch *et al*. 1985). PTX treatment similarly attenuated SRIF action in GH_4 cells overexpressing sst2 (Tentler *et al*. 1997). In addition, PTX attenuated SRIF-dependent inhibition of GHRH-induced GH in MtT/SGL somatotroph cells (Morishita *et al*. 2003). sst5-specific inhibition of forskolin-stimulated cAMP accumulation was dependent on $G\alpha_{o1}$ in human GH-secreting tumor primary cultures (Peverelli *et al*. 2013). In AtT-20 cells, SRIF inhibition of adenylate cyclase and ACTH secretion is $G\alpha_{i1}$-dependent (Tallent & Reisine 1992). sst2, sst3, and sst5 mediate SRIF-dependent inhibition of cAMP in AtT-20 cells (Ben-Shlomo *et al*. 2005), as does sst1 in GC cells (Cervia *et al*. 2003). In chicken pituitary cells, SRIF inhibited GHRH-induced GH release by inhibiting cAMP/PKA signaling independent of calcium or protein kinase C (Donoghue & Scanes 1991). Although SRIF is classically an inhibitor of adenylate cyclase activity, there are reports using primary porcine somatotroph cultures in which both low and high doses of SRIF increased cAMP levels, suggesting a possible dual dose-dependent effect (Ramirez *et al*. 2002). As the mechanism by which this phenomenon occurs remains elusive, perhaps SSTRs, such as other GPCRs, might interact with not only $G\alpha_{i/o}$ proteins but also $G\alpha_s$ depending on the ligand context and receptor conformation. Therefore, SRIF-mediated inhibition of pituitary hormone secretion is both Ca^{2+} and cAMP dependent; as both Ca^{2+} concentration and cAMP levels are $G\alpha_i$ dependent, it is difficult to determine the relative contribution of each to hormone secretion and whether these mechanisms occur independently of each other.

Protein phosphatase pathways

SRIF-mediated regulation of pituitary protein phosphatase pathways is primarily associated with mechanisms controlling cell growth. SRIF is associated with increased protein phosphatase activity in both human GH-secreting pituitary adenoma cells and rat cell lines as well as human non-functioning pituitary tumors (Cervia & Bagnoli 2007). SRIF increases tyrosine phosphatase activity and was associated with the inhibition of cell growth in human GH-secreting pituitary tumor cells *in vitro* (Florio *et al*. 2003), while serine/threonine phosphatase activity participates in SRIF-mediated regulation of Ca^{2+} influx through dephosphorylation of Ca^{2+} and K^+ voltage-activated (BK) channels (White *et al*. 1991). Octreotide exhibited antiproliferative effects in GH_3 cells, mediated by both PTX-dependent and SHP1

http://jme.endocrinology-journals.org
DOI: 10.1530/JME-14-0034

Published by Bioscientifica Ltd.

(but not SHP2)-dependent mechanism (Theodoropoulou *et al*. 2006, Cerovac *et al*. 2010). Octreotide induced SHP1-dependent inhibition of the PI3K activity leading to the inhibition of PDK1 and Akt activity, ultimately leading to enhanced glycogen synthase kinase 3β (GSK3β) activity and upregulation of the tumor suppressor *ZAC1* (*ZACN*; Theodoropoulou *et al*. 2006). Octreotide also increased the levels of rapamycin-suppressed phosphorylated insulin receptor substrate 1, subsequently decreasing phosphorylation of Akt through SHP1 (Cerovac *et al*. 2010). Interestingly, in acromegaly patients treated with octreotide, *ZAC1* immunoreactivity correlated with insulin-like growth factor 1 (IGF1) normalization and tumor shrinkage (Theodoropoulou *et al*. 2009). Both octreotide and an sst2-specific agonist (BIM23120) induced apoptosis, and apoptosis-associated gene expression in human GH-secreting tumors is blocked by vanadate, indicating the involvement of protein phosphatases (Ferrante *et al*. 2006). Vanadate similarly inhibited SRIF and lanreotide-induced growth arrest in primary cultures of non-functioning pituitary adenomas (Florio *et al*. 1999). The role of SRIF-mediated regulation of protein phosphatase activity and growth arrest in non-pituitary cells has been described (Florio 2008). As, clinically, SRIF analog therapy induces pituitary tumor shrinkage, the mechanisms involved in SRIF-mediated protein phosphatase activation require further investigation. Moreover, the contribution of individual SSTR subtypes to phosphatase activation is still unclear.

Other SRIF-dependent pituitary pathways

Several other signaling pathways have been described to mediate SRIF action (Cervia & Bagnoli 2007, Otsuka *et al*. 2012), including MAPK, guanylyl cyclase, PKC, nitric oxide (NO), PI3K/Akt, and bone morphogenetic proteins (BMPs). The physiological relevance of these pathways to pituitary cell growth and hormone secretion remains unclear. Both octreotide and pasireotide decreased MAPK/ERK phosphorylation in both GH_3 cells and in GH-secreting tumor cultures, and upregulated $p27^{Kip}$ expression (Hubina *et al*. 2006), while knockdown of sst5 increased ERK phosphorylation in AtT-20 cells (Ben-Shlomo *et al*. 2007). In human GH-secreting tumor cultures, sst5-dependent inhibition of ERK phosphorylation was dependent on $G\alpha_{o1}$ (Peverelli *et al*. 2013). Octreotide activated PI3K/Akt and MAPK pathways through sst2 and sst5 in GH-secreting cells, inducing histone methyltransferases associated with menin, resulting in the upregulation of $p27^{Kip}$ and cell cycle arrest (Horiguchi *et al*. 2009). These discrepancies may suggest cell-type specificity or perhaps a similar dual role for SRIF action as that for adenylate cyclase regulation. SRIF was also shown to exhibit a dual dose-dependent effect on pituitary guanylyl cyclase regulation and cGMP accumulation (Vesely 1980).

SRIF inhibited both PKC-induced stimulation of GH secretion (Ikuyama *et al*. 1987) and NO-induced cGMP and GH levels (Bocca *et al*. 2000, Luque *et al*. 2005). SRIF blocked phospholipase A2-mediated GHRH- and thyrotropin-releasing hormone (TRH)-induced pituitary arachidonate release (Judd *et al*. 1986) and decreased arachidonate levels in GC cells (Cervia *et al*. 2002*b*). SRIF was also shown to induce apoptosis through NFκB/JNK/caspase pathway (Ferrante *et al*. 2006, Guillermet-Guibert *et al*. 2007), and inhibited vascular endothelial growth factor production levels in pituitary tumor cells (Lohrer *et al*. 2001, Zatelli *et al*. 2007). SRIF antagonized CRH-dependent inhibition of GSK3β activity in AtT-20 cells, inhibiting Wnt/β-catenin-mediated transcription and cell growth in a cAMP-dependent manner (Khattak *et al*. 2010). SRIF action in AtT-20 cells is dependent on BMP signaling. Inhibitory effects of octreotide and pasireotide on CRH-induced secretion in AtT-20 cells were attenuated by noggin, an inhibitory BMP-binding protein, suggesting that the endogenous BMP system is functionally linked to the mechanism of SRIF-mediated inhibition of secretion (Tsukamoto *et al*. 2010). SRIF-signaling in GH_3 cells was similarly shown to be dependent on BMPs (Tsukamoto *et al*. 2011). Although it is evident that kinases and BMP growth factors are involved in pituitary SRIF signaling, our understanding of their regulation and functional consequences remain unclear.

Receptor phosphorylation, internalization, and desensitisation

Following activation of SSTRs by ligand, a feedback mechanism is activated leading to the receptor phosphorylation and internalization, ultimately initiating receptor desensitization and attenuated receptor-related signaling. As of yet, the only SSTR subtype described to follow this process in pituitary cells is sst2. sst2 was phosphorylated and internalized after treatment with SRIF and sst2-specific agonists in stable sst2 transfectant GH4C1 cells (Hipkin *et al*. 1997). sst2 is phosphorylated at five serine and threonine residues within the C-terminus following SRIF treatment (Liu *et al*. 2009). Moreover, in transiently transfected GH_3 cells, both SRIF and octreotide, but not pasireotide, induced robust sst2 phosphorylation. Pasireotide stimulated selective residue

http://jme.endocrinology-journals.org
DOI: 10.1530/JME-14-0034

Published by Bioscientifica Ltd.

phosphorylation only upon GRK2 and GRK3 overexpression, yet resulted in only weak β-arrestin–sst2 complexes that easily dissociated (Poll *et al*. 2010). Prolonged SRIF stimulation leads to sst2 desensitization in both GH_4C_1 and AtT-20 cells (Hipkin *et al*. 1997), leading to attenuated responses to SRIF inhibition of cAMP production, and enhanced forskolin and CRH induction of adenylate cyclase and cAMP levels (Reisine & Axelrod 1983, Presky & Schonbrunn 1988, Ben-Shlomo *et al*. 2009).

While sst1 does not internalize (Sarret *et al*. 1999), there are conflicting reports regarding sst5 internalization. One study using transient sst5 transfectant GH_3 cells found that the third intracellular loop of sst5 was involved in receptor phosphorylation and internalization following β-arrestin 2 binding (Peverelli *et al*. 2008). Yet, sst5 did not internalize following treatment with either SRIF or sst5-specific agonists in stable AtT-20 transfectants (Sarret *et al*. 1999, Ben-Shlomo *et al*. 2005). SRIF desensitization in stable receptor transfectant AtT-20 cells was dependent on sst2 and not sst5, as sst5 did not internalize in these cells (Ben-Shlomo *et al*. 2009). sst5 expression is unaffected by sst5 agonists or pasireotide in AtT-20 cells (van der Hoek *et al*. 2005, Ben-Shlomo *et al*. 2009), suggesting that sst5 remains biologically active in the membrane longer than sst2. *In vitro* studies may not accurately represent patterns of SRIF-dependent SSTR subtype-specific internalization and desensitization *in vivo* and therefore requires further study.

Any pituitary cell may express multiple sst receptor subtypes on the cell surface (Ben-Shlomo & Melmed 2010), suggesting that in addition to receptor subtype-specific signaling, receptor subtypes may form heterodimers, which may govern pituitary cell response to SRIF and SRIF analogs. Although sst receptor dimerization has yet to be demonstrated in pituitary cells lines, there is evidence of sst receptor hetero-dimerization when receptors are stably overexpressed in non-pituitary cell lines, and that dimerization does effect sst receptor function (Pfeiffer *et al*. 2002, Grant *et al*. 2008, War & Kumar 2012). While sst receptor subtypes are structurally and sequentially conserved throughout the body, it is likely that variation in receptor subtype expression contributes to the tissue specificity of SRIF-mediated molecular signaling, and should be further evaluated.

SRIF-dependent pituitary hormone secretion

SRIF exerts a primary effect on pituitary cells through acute inhibition of hormone secretion, specifically by suppressing exocytosis of hormone-containing vesicles. Although, it remains unclear whether SRIF-dependent inhibition of secretion is partially contingent on inhibition of cell growth arrest, as they usually occur concurrently following SRIF analog treatment of somatotroph tumors, there are reports of asynchronous hormone secretion and tumor shrinkage (Colao *et al*. 2009). In one such tumor, expression of sst5 was higher than sst2 levels, suggesting the potential of subtype-specific regulation of hormone secretion vs antiproliferative effects (Resmini *et al*. 2007).

Although SRIF is the primary inhibitor of pituitary GH secretion and is the main focus of this review, it should be noted that the neuropeptide CST, which shares structural homology to SRIF, has been reported to exhibit a similar inhibitory effect as SRIF on GH secretion both *in vitro* and *in vivo* (Broglio *et al*. 2002), yet the mechanism of action remains unclear as CST is reported to bind not only to SSTRs but also to the ghrelin receptor (GHS-R; Broglio *et al*. 2002). In contrast to the inhibitory role of CST on GH secretion, CST is reported to increase PRL release (Baranowska *et al*. 2009, Cordoba-Chacon *et al*. 2011). Unlike SRIF, CST has not been shown to be secreted from the hypothalamus directly into the pituitary portal system, and the exact role of CST in endocrine regulation of pituitary function *in vivo* is yet to be demonstrated.

GH secretion

The classical outcome of SRIF/sst signaling is the inhibition of hormone secretion, particularly that of GH from pituitary somatotroph cells (Giustina & Veldhuis 1998). Multiple factors and feedback loops regulate the release of SRIF from the hypothalamus and ultimately the control of GH secretion, including serum GH/IGF1 and glucose, as well as immobilization and exercise (Giustina & Veldhuis 1998). As SRIF inhibits GHRH-induced GH transcription and secretion by suppressing exocytosis of hormone-containing granules (Patel & Srikant 1986, Tentler *et al*. 1997, Morishita *et al*. 2003, Farhy & Veldhuis 2004), *SRIF*-null mice were expected to exhibit the characteristics of GH excess. However, despite moderately elevated GH levels, serum IGF1 levels were slightly elevated; however, body length and weight and IGF1 levels were unchanged (Low *et al*. 2001, Zeyda *et al*. 2001). Targeted hypothalamic delivery of lentiviral-shRNA against SRIF in young mice led to increased GH protein levels without effecting GH mRNA levels, yet serum levels of SRIF, GH, and IGF1, as well as body weight, were unchanged (Hao *et al*. 2010). These studies suggest that although SRIF may play an important function as an acute inhibitor of GHRH-induced

http://jme.endocrinology-journals.org
DOI: 10.1530/JME-14-0034

Published by Bioscientifica Ltd.

GH secretion, it may have a less significant role in regulation of basal GH secretion.

While direct inhibition of pituitary hormone transcription has yet to be definitively associated with SRIF signaling, as some studies demonstrate a SRIF-dependent decrease in GH mRNA levels (Sugihara *et al*. 1993, Tsukamoto *et al*. 1994, Acunzo *et al*. 2008) others describe no change (Simard *et al*. 1986, Davis *et al*. 1989, Namba *et al*. 1989, Tanner *et al*. 1990, Gruszka *et al*. 2007), and some even demonstrate upregulation of gene expression possibly reflecting a GH-rebound effect after termination of SRIF treatment; the latter exemplifies the importance of outcome measurement timing. SRIF did not affect GH mRNA expression, but did suppress intracellular GH protein levels and decreased GH secretion in primary rat anterior pituitary cells (Simard *et al*. 1986). Similarly, SRIF inhibited GH secretion without affecting *GH* mRNA expression in primary human GH-secreting tumors cells (Davis *et al*. 1989). In cultured bovine pituitary cells, SRIF was able to suppress GHRH-induced GH expression, but had no effect on untreated cells (Tanner *et al*. 1990). Moreover, while sst2-specific and sst5-specific agonists suppressed GH secretion in human GH-secreting tumors, they did not affect GH transcription (Gruszka *et al*. 2012). Nevertheless, transient overexpression of sst2 in primary human GH secreting tumor cells modestly suppressed GH expression, in the absence of SRIF ligand (Acunzo *et al*. 2008).

Delineation of sst subtype-selective SRIF-mediated regulation of GH secretion remains unclear; however, both sst2 and sst5 and to a lesser extent sst1 play important roles in the inhibition of GH secretion. SRIF analog therapies targeting sst2 and/or sst5 for treatment of patients with GH-secreting pituitary tumors are effective at reducing serum GH and normalizing serum IGF1 levels (Melmed 2006). Moreover, treatment using compounds with affinity for sst2 and sst5 was 40% more effective at suppressing primary GH-secreting tumor GH secretion than sst2 or sst5-selective agonists individually (Shimon *et al*. 1997*a*). Increased sst2 membrane density in GH-secreting tumor cells enhances the sensitivity to sst2-selective agonists (Acunzo *et al*. 2008, Taboada *et al*. 2008). In normal fetal pituitary, both sst2 and sst5-specific agonists inhibit GHRH-induced GH secretion (Shimon *et al*. 1997*b*, Ren *et al*. 2003); however, co-treatment with sst2 and sst5-selective agonists was more effective than each individually (Ren *et al*. 2003). sst1-selective agonists similarly reduced GH secretion in both primary tumor cultures (Zatelli *et al*. 2003) as well as GC cells (Cervia *et al*. 2002*a*). Pasireotide, which binds all four receptor subtypes (sst5 > sst2 > sst3 > sst1), more effectively reduces serum GH levels in animal models compared with octreotide (sst2 > sst5 ⋙ sst3, but not sst1), and was recently demonstrated to have some advantage over octreotide in patients with acromegaly (Colao *et al*. 2014); however, the long-term efficacy of one drug over the other is yet to be proven (Petersenn *et al*. 2014*a*,*b*) and is still under current investigation.

Importantly, recent reports demonstrate a concentration-dependent, cell-specific effect of SRIF on GH-secreting cells, in both bovine and primates. While high-concentrations of SRIF inhibit GH-secretion, low-concentrations stimulate GH-secretion. sst1 and sst2 were shown to mediate the inhibitory effect of SRIF, while its stimulatory effect was signaled via sst5, all through adenylate cyclase–cAMP pathway and intracellular calcium level regulation. SRIF's dose-dependent stimulatory/inhibitory effects should be further studied as they may have an important role in the physiological regulation of somatotroph cells (Luque *et al*. 2006*a*, Cordoba-Chacon *et al*. 2012).

Surprisingly, despite the dominant role of sst2 and sst5 in SRIF-mediated inhibition of GHRH-induced GH secretion, *Sst2* and *Sst5*-null mice do not exhibit elevated serum GH levels (Zheng *et al*. 1997, Norman *et al*. 2002, Luque *et al*. 2006*b*). These results are, however, consistent with a potential, yet unproven, compensatory function of sst1 or sst3 regulating GH secretion in the absence of sst2 and sst5. Conditional knockout mice may provide a model to further elucidate the subtype-specific role of sst receptor subtypes in SRIF-dependent inhibition of GH secretion.

PRL secretion

SRIF-dependent control of PRL secretion is modest compared with that of GH. Treatment of prolactinoma samples with sst5-selective agonists effectively inhibited PRL secretion without affecting PRL expression (Shimon *et al*. 1997*a*, Jaquet *et al*. 1999, Fusco *et al*. 2008, Gruszka *et al*. 2012), while octreotide had no effect, likely due to the fact that sst5 is the predominantly expressed receptor subtype in human prolactinoma samples (Jaquet *et al*. 1999). Similarly, while octreotide exerted only modest effects, pasireotide strongly suppressed PRL secretion in prolactinoma tumor cultures (Hofland *et al*. 2004). PRL secretion was reduced upon SRIF treatment as well as an sst1-specific agonist in PRL-secreting pituitary tumors; the degree of PRL suppression correlated with sst1 expression levels (Zatelli *et al*. 2003). BMP4 enhanced the attenuating effect of pasireotide, but not of octreotide, on

http://jme.endocrinology-journals.org
DOI: 10.1530/JME-14-0034

Published by Bioscientifica Ltd.

forskolin-induced PRL in GH_3 cells. Interestingly, BMP4 and BMP6 downregulated endogenous sst2 abundance, while increasing sst5 expression, and treatment with noggin rescued these effects. Moreover, noggin treatment increased octreotide sensitivity and decreased pasireotide sensitivity (Tsukamoto *et al*. 2011).

Several studies suggest that SRIF-dependent regulation of PRL secretion is estrogen-dependent. Female rats pretreated with 17β-estradiol (E_2) exhibited lanreotide-dependent reduction in lactotroph cell density and PRL secretion (Schussler *et al*. 1994). E_2 treatment sensitized PRL-secreting tumor cells to SRIF and octreotide-dependent inhibition of PRL secretion, likely due to the upregulation of sst2 and sst3 (Visser-Wisselaar *et al*. 1997, Djordjijevic *et al*. 1998). Despite the inability of SRIF to inhibit PRL secretion in male rat primary lactotroph cultures, E_2 treatment similarly sensitizes these cells to SRIF (Goth *et al*. 1996, Lee & Shin 1996). In addition, SRIF inhibits PRL induction by estrogen in male-to-female transsexuals, even more so upon co-treatment with cyproterone acetate, a compound with anti-androgen characteristics (Gooren *et al*. 1984). Taken together, the regulation of PRL secretion through SRIF-dependent pathways appears to be receptor subtype specific, BMP dependent, and sensitive to the presence of estrogen.

ACTH secretion

The role of SRIF signaling in the regulation of ACTH secretion from pituitary corticotroph cells remains unclear. SRIF did not affect basal or CRH (Stafford *et al*. 1989), ghrelin (Broglio *et al*. 2002), or angiotensin II (Volpi *et al*. 1996)-stimulated ACTH or cortisol levels in humans. Nevertheless, studies suggest that SRIF regulates ACTH secretion and is dependent on cortisol levels and cell milieu (Hofland 2008). Although SRIF did not affect basal or CRH-stimulated ACTH secretion in normal rat pituitary cells (Brown *et al*. 1984, Kraicer *et al*. 1985), SRIF inhibited CRH- and vasopressin-induced ACTH secretion in cultured pituitary cells derived from adrenalectomized rats and in serum starved cultures (Hofland 2008). Increasing cortisol levels in cell medium downregulated sst2 but not sst5 expression in corticotroph tumor cells that express both receptors (van der Hoek *et al*. 2007, van der Pas *et al*. 2013). Pituitary corticotroph cells were sensitized to octreotide in a serum-free environment, as well as after inhibition of the glucocorticoid receptor, which also downregulates sst2 expression (Lamberts *et al*. 1989*a*).

Octreotide and lanreotide, both clinically used as sst2 agonists, were ineffective in treating patients with Cushing's disease (Hofland 2008), but were able to suppress ACTH levels in patients with hypercortisol-emia such as those with adrenal insufficiency (Fehm *et al*. 1976) and Nelson's syndrome (Tyrrell *et al*. 1975, Lamberts *et al*. 1989*b*). However, pasireotide, which has preferential affinity for sst5, inhibits ACTH secretion in patients harboring ACTH-secreting adenoma despite hypercortisolemia (Colao *et al*. 2012). The significance of SRIF-mediated signaling, particularly through sst5, has been further delineated through investigation of *Srif*-null and *Sst5*-null mice models; *Srif*-null mice exhibit elevated levels of *Pomc* mRNA expression (Luque *et al*. 2006*b*), and sst5-null mice have increased basal serum ACTH and cortisol levels (Strowski *et al*. 2003).

Importantly, downregulation of sst2 may not be the sole explanation to octreotide resistance as the drug was less effective than pasireotide at reducing ACTH secretion even after normalization of cortisol levels and rescued sst2 expression in preoperative Cushing's patients (van der Pas *et al*. 2013). Therefore the contribution of other SSTRs expressed on corticotroph cells including sst3 and sst1 may play a role as well.

BMP signaling was shown to play a significant role in sst receptor-mediated inhibition of ACTH secretion in corticotroph cells. The BMP inhibitor noggin enhances CRH-induced ACTH secretion in AtT-20 cells and attenuated octreotide and pasireotide-mediated suppression of CRH-induced ACTH secretion. Octreotide and pasireotide increased BMP–Smad1/5/8 signaling and upregulated BMP type I and II receptors while simultaneously downregulating inhibitory Smad6/7 (Tsukamoto *et al*. 2010). In summary, SRIF-mediated regulation of ACTH secretion appears to be sst receptor subtype specific and dependent on serum cortisol levels. The role of sst1, sst3, and BMP signaling pathways in the regulation of ACTH secretion require further investigation.

Gonadotropin secretion

Knowledge of SRIF regulation of human LH and FSH secretion is limited as pituitary tumors arising from gonadotroph lineages do not usually secrete FSH or LH and therefore do no result in a phenotype associated with hormone hypersecretion (Greenman & Stern 2009). Nevertheless, evidence suggests an inhibitory effect of SRIF on gonadotropin secretion. SRIF infusion suppressed gonadotropin-releasing hormone (GNRH)-induced LH and FSH in normal men (Millar *et al*. 1982) and inhibited LH pulse amplitude, but not frequency, without affecting FSH pulsatility (Samuels *et al*. 1992). SRIF does not affect

http://jme.endocrinology-journals.org
DOI: 10.1530/JME-14-0034

Published by Bioscientifica Ltd.

basal secretion of LH or FSH in cultured pituitaries from male rats (Yu *et al*. 1997), yet suppresses GNRH-induced LH but not FSH (Yu *et al*. 1997, Starcevic *et al*. 2002). In addition, SRIF suppressed gonadotropin levels in 60% of FSH-producing pituitary tumors and 30% of LH-secreting pituitary adenoma cultures (Klibanski *et al*. 1991).

TSH secretion

Although SRIF inhibits TSH secretion, the effect is less pronounced than that of GH secretion from somatotrophs (Patel & Srikant 1986). Both sst2 and sst5 were implicated in the suppression of TSH secretion (Shimon *et al*. 1997*b*); however, the relative contribution of individual receptor subtypes remains unknown. SRIF inhibited TRH-induced TSH secretion in normal adult males (Spoudeas *et al*. 1992). Similarly, SRIF suppressed TSH pulse amplitude and frequency (Samuels *et al*. 1992) and inhibited TSH levels in normal volunteers and in patients with primary hypothyroidism (Reichlin 1983). Octreotide and lanreotide reduced TSH secretion and normalized free thyroxine and free tri-iodothyronine levels in patients harboring pituitary TSH-secreting adenomas (Gancel *et al*. 1994, Beck-Peccoz & Persani 2002). Similarly, octreotide suppressed serum TSH concentrations in nine patients harboring TSH-secreting tumors, and similarly suppressed TSH secretion in cell cultures (Bertherat *et al*. 1992). TSH-secreting adenomas are extremely rare, limiting the ability to comprehensively study sst receptor subtype-specific regulation of TSH secretion.

In summary, SRIF is a dominant inhibitor of both basal and induced pituitary hormone secretion. There are clear indications that sst receptor-signaling pathway activation is receptor subtype- and density-specific, as well as cell type and context specific. While GH secretion is mediated predominantly through sst2, and to a lesser extent sst5, ACTH and PRL secretion appear to be coordinated for the most part through sst5 signaling. Further study is required to delineate SRIF receptor subtype specificity and to elucidate the role of sst1 and sst3 in pituitary hormone secretion.

SRIF-independent constitutive sst receptor activity

Constitutive receptor activity is the ability of a receptor to adopt an active conformation independently from its selective agonist (Seifert & Wenzel-Seifert 2002). Multiple GPCRs exhibit constitutive activity in their WT form and some by acquiring naturally occurring disease-causing mutations (Seifert & Wenzel-Seifert 2002).

Partial knockdown of sst2, sst3, or sst5 in mouse ACTH-secreting pituitary AtT-20 cells resulted in increased baseline intracellular cAMP levels and consequently ACTH secretion (Ben-Shlomo *et al*. 2007), while overexpression of either sst2 or sst5 in these cells resulted in reduced cellular response CRH via downregulation of CRH receptor subtype 1 (CRH-R1) expression (Ben-Shlomo *et al*. 2009). In addition, moderate sst2 overexpression in rat GH-secreting pituitary tumor cells (GC cell line) resulted in significantly decreased GH synthesis partially via GH promoter de-acetylation, which was not observed when a sst2 DRY-motif mutant lacking constitutive activity (Ben-Shlomo *et al*. 2013) was stably overexpressed. Inhibition of GH transcription was also observed when human pituitary cell primary cultures were infected with a low dose of sst2-containing adenovirus (Acunzo *et al*. 2008).

Utilizing a similar approach to study sst3 in GH-secreting cells, we show that stable sst3 transfectants exhibited suppressed basal intracellular cAMP levels, PKA activity, and inhibition of GH transcription, though to a lesser extent as compared with sst2 overexpression. sst3-mediated GH inhibition was not regulated epigenetically but rather via dephosphorylation and thus activation of GSK3β, a PKA substrate. The cells expressing non-constitutively active sst3 mutated at its DRY motif were unaffected (Eigler *et al*. 2014).

Constitutive sst receptor activity is yet to be proven *in vivo*. This is challenging as a naturally occurring, disease-causing, constitutively active SSTR mutant has not yet been characterized, and an SSTR inverse agonist is not available. Moreover, the SRIF system exhibits significant redundancy, as CST and SRIF bind all sst receptor subtypes with similar affinities and the receptors also share multiple signaling pathways. To rigorously study constitutive sst2 activity *in vivo* will require an animal experimental model that expresses neither ligands nor all other sst receptor subtypes, a difficult task.

Evidence in the literature suggests the possible presence of constitutive sst2 activity. Intriguing evidence from mice points to conditions in which SRIF (not sst2) is dispensable for determining baseline GH control. First, abolishing SRIF-producing rat hypothalamic neurons resulted in acutely increased serum GH levels, which normalized within 10 days, without altered pituitary GH-content (Soya & Suzuki 1990). Second, knockout *Srif* $^{(-/-)}$, *Cort* $^{(-/-)}$, and double-knockout *Srif* $^{(-/-)}$/*Cort* $^{(-/-)}$ mice do not exhibit excessive growth

http://jme.endocrinology-journals.org
DOI: 10.1530/JME-14-0034

Published by Bioscientifica Ltd.

(de Lecea and Castano 2006, Zeyda & Hochgeschwender 2008, Cordoba-Chacon *et al.* 2011). In addition, hypersomatostatinemia elaborated by abdominal somatostatinoma tumors is not associated with GH deficiency (Oberg & Eriksson 2005, Galli *et al.* 2006). Observed negative association between sst2 and GH expression support the existence of a biological relationship between the two. For example, *Srif*$^{(-/-)}$ mice exhibit 1.5- to threefold increase in GH levels along with 70% decrease in sst2 levels (Low *et al.* 2001, Zeyda *et al.* 2001). Glucocorticoids, acutely downregulate *Sst2* promoter activity associated with increased pituitary GH synthesis in SRIF-free conditions (Xu *et al.* 1995, Zeyda *et al.* 2001, Kajimura *et al.* 2003). In contrast, adrenalectomy (cortisol deficiency) increased rat somatotroph sst2 levels (Hofland 2008). In addition, E_2 lowers sst2 levels and increases baseline GH in the absence or presence of SRIF (Cardenas *et al.* 2003, Borghi *et al.* 2006, Elango *et al.* 2006). Aromatase-null (i.e. E_2 deficient) female mice exhibited high pituitary *Sst2* gene expression with concomitant low GH levels, all reversed with E_2 treatment (Yan *et al.* 2004). Importantly, female rats exhibit continuous GH secretion with higher baseline levels and also have lower *Sst2* expression as compared with males, while male rats treated with E_2 exhibited increased baseline GH and downregulation of sst receptors (Baumeister & Meyerhof 2000). In summary, when sst2 is decreased, GH is increased, with or without SRIF. Although intriguing, the physiological importance of constitutive sst receptor activity is yet unclear *in vivo*. As absolute receptor number in the cell also determines observed constitutive activity level, receptor expression level regulation by SRIF and other factors may control constitutive sst receptor activity.

Conclusion

SRIF system, i.e. hypothalamic SRIF14 and its cognate sst1, sst2, sst3, and sst5 receptor subtypes, control pituitary gland function, mostly inhibiting anterior pituitary gland basal and induced hormone secretion. SRIF–sst receptor signaling pathway activation is receptor subtype specific and density specific, as well as cell type and context specific. SRIF/sst2 is the main mediator of GH secretion while SRIF/sst5 mainly mediates ACTH and PRL secretion. Sst receptor activation mediates its effect through multiple pathways, mainly adenylate cyclase/cAMP/PKA, MAPK, and ion channel regulation. SRIF-independent constitutive sst receptor activity is present in pituitary cells *in vitro*, inhibiting cellular cAMP and ACTH responses to CRH and GH transcription.

Declaration of interest

The authors declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of the review.

Funding

This review did not receive any specific grant from any funding agency in the public, commercial or not-for-profit sector.

References

Abe H, Chihara K, Minamitani N, Iwasaki J, Chiba T, Matsukura S & Fujita T 1981 Stimulation by bombesin of immunoreactive somatostatin release into rat hypophysial portal blood. *Endocrinology* **109** 229–234. (doi:10.1210/endo-109-1-229)

Acunzo J, Thirion S, Roche C, Saveanu A, Gunz G, Germanetti AL, Couderc B, Cohen R, Figarella-Branger D, Dufour H *et al.* 2008 Somatostatin receptor sst2 decreases cell viability and hormonal hypersecretion and reverses octreotide resistance of human pituitary adenomas. *Cancer Research* **68** 10163–10170. (doi:10.1158/0008-5472.CAN-08-1857)

Aguila MC 1998 Somatostatin decreases somatostatin messenger ribonucleic acid levels in the rat periventricular nucleus. *Peptides* **19** 1573–1579. (doi:10.1016/S0196-9781(98)00109-0)

Baranowska B, Bik W, Baranowska-Bik A, Wolinska-Witort E, Chmielowska M & Martynska L 2009 Cortistatin and pituitary hormone secretion in rat. *Journal of Physiology and Pharmacology* **60** 151–156.

Bauer W, Briner U, Doepfner W, Haller R, Huguenin R, Marbach P, Petcher TJ & Pless 1982 SMS201-995: a very potent and selective octapeptide analogue of somatostatin with prolonged action. *Life Sciences* **31** 1133–1140. (doi:10.1016/0024-3205(82)90087-X)

Baumeister H & Meyerhof W 2000 Gene regulation of somatostatin receptors in rats. *Journal of Physiology* **94** 167–177. (doi:10.1016/S0928-4257(00)00201-1)

Beck-Peccoz P & Persani L 2002 Medical management of thyrotropin-secreting pituitary adenomas. *Pituitary* **5** 83–88. (doi:10.1023/A:1022360414062)

Ben-Shlomo A & Melmed S 2010 Pituitary somatostatin receptor signaling. *Trends in Endocrinology and Metabolism* **21** 123–133. (doi:10.1016/j.tem.2009.12.003)

Ben-Shlomo A, Wawrowsky KA, Proekt I, Wolkenfeld NM, Ren SG, Taylor J, Culler MD & Melmed S 2005 Somatostatin receptor type 5 modulates somatostatin receptor type 2 regulation of adrenocorticotropin secretion. *Journal of Biological Chemistry* **280** 24011–24021. (doi:10.1074/jbc.M501998200)

Ben-Shlomo A, Pichurin O, Barshop NJ, Wawrowsky KA, Taylor J, Culler MD, Chesnokova V, Liu NA & Melmed S 2007 Selective regulation of somatostatin receptor subtype signaling: evidence for constitutive receptor activation. *Molecular Endocrinology* **21** 2565–2578. (doi:10.1210/me.2007-0081)

Ben-Shlomo A, Zhou C, Pichurin O, Chesnokova V, Liu NA, Culler MD & Melmed S 2009 Constitutive somatostatin receptor activity determines tonic pituitary cell response. *Molecular Endocrinology* **23** 337–348. (doi:10.1210/me.2008-0361)

Ben-Shlomo A, Pichurin O, Khalafi R, Zhou C, Chesnokova V, Ren SG, Liu NA & Melmed S 2013 Constitutive somatostatin receptor subtype 2 activity attenuates GH synthesis. *Endocrinology* **154** 2399–2409. (doi:10.1210/en.2013-1132)

Berelowitz M, Maeda K, Harris S & Frohman LA 1980 The effect of alterations in the pituitary-thyroid axis on hypothalamic content and *in vitro* release of somatostatin-like immunoreactivity. *Endocrinology* **107** 24–29. (doi:10.1210/endo-107-1-24)

http://jme.endocrinology-journals.org
DOI: 10.1530/JME-14-0034

Published by Bioscientifica Ltd.

Berelowitz M, Szabo M, Frohman LA, Firestone S, Chu L & Hintz RL 1981 Somatomedin-C mediates growth hormone negative feedback by effects on both the hypothalamus and the pituitary. *Science* **212** 1279–1281. (doi:10.1126/science.6262917)

Berelowitz M, Dudlak D & Frohman LA 1982 Release of somatostatin-like immunoreactivity from incubated rat hypothalamus and cerebral cortex. Effects of glucose and glucoregulatory hormones. *Journal of Clinical Investigation* **69** 1293–1301. (doi:10.1172/JCI110569)

Berelowitz M, Xu Y, Song J & Bruno JF 1995 Regulation of somatostatin receptor mRNA expression. *Ciba Foundation Symposium* **190** 111–122; discussion 122-116.

Bertherat J, Brue T, Enjalbert A, Gunz G, Rasolonjanahary R, Warnet A, Jaquet P & Epelbaum J 1992 Somatostatin receptors on thyrotropin-secreting pituitary adenomas: comparison with the inhibitory effects of octreotide upon *in vivo* and *in vitro* hormonal secretions. *Journal of Clinical Endocrinology and Metabolism* **75** 540–546. (doi:10.1210/jcem.75.2.1353505)

Bichet DG 2011 The Posterior pituitary. In *Pituitary*, pp 261–299. Ed S Melmed. San Diego, California: Elsevier.

Bilezikjian LM & Vale WW 1983 Stimulation of adenosine 3′,5′-monophosphate production by growth hormone-releasing factor and its inhibition by somatostatin in anterior pituitary cells *in vitro*. *Endocrinology* **113** 1726–1731. (doi:10.1210/endo-113-5-1726)

Bjoro T, Ostberg BC, Sand O, Gordeladze J, Iversen JG, Torjesen PA, Gautvik KM & Haug E 1987 Vasoactive intestinal peptide and peptide with N-terminal histidine and C-terminal isoleucine increase prolactin secretion in cultured rat pituitary cells (GH4C1) via a cAMP-dependent mechanism which involves transient elevation of intracellular Ca^{2+}. *Molecular and Cellular Endocrinology* **49** 119–128. (doi:10.1016/0303-7207(87)90205-X)

Bocca L, Valenti S, Cuttica CM, Spaziante R, Giordano G & Giusti M 2000 Nitric oxide biphasically modulates GH secretion in cultured cells of GH-secreting human pituitary adenomas. *Minerva Endocrinologica* **25** 55–59.

Bonnefont X, Fiekers J, Creff A & Mollard P 2000 Rhythmic bursts of calcium transients in acute anterior pituitary slices. *Endocrinology* **141** 868–875. (doi:10.1210/endo.141.3.7363)

Borghi MM, Longui CA, Calliari LE, Faria CD, Kochi C & Monte O 2006 Transdermal estradiol priming during clonidine stimulation test in non-growth hormone deficient children with short stature: a pilot study. *Journal of Pediatric Endocrinology & Metabolism* **19** 223–227. (doi:10.1515/JPEM.2006.19.3.223)

Broglio F, Arvat E, Benso A, Gottero C, Prodam F, Grottoli S, Papotti M, Muccioli G, van der Lely AJ, Deghenghi R *et al.* 2002 Endocrine activities of cortistatin-14 and its interaction with GHRH and ghrelin in humans. *Journal of Clinical Endocrinology and Metabolism* **87** 3783–3790. (doi:10.1210/jcem.87.8.8749)

Brown MR, Rivier C & Vale W 1984 Central nervous system regulation of adrenocorticotropin secretion: role of somatostatins. *Endocrinology* **114** 1546–1549. (doi:10.1210/endo-114-5-1546)

Bruns C, Lewis I, Briner U, Meno-Tetang G & Weckbecker G 2002 SOM230: a novel somatostatin peptidomimetic with broad somatotropin release inhibiting factor (SRIF) receptor binding and a unique antisecretory profile. *European Journal of Endocrinology* **146** 707–716. (doi:10.1530/eje.0.1460707)

Burgus R, Ling N, Butcher M & Guillemin R 1973 Primary structure of somatostatin, a hypothalamic peptide that inhibits the secretion of pituitary growth hormone. *PNAS* **70** 684–688. (doi:10.1073/pnas.70.3.684)

Canosa LF, Lin X & Peter RE 2003 Effects of sex steroid hormones on the expression of somatostatin receptors sst1 and sst5 in goldfish pituitary and forebrain. *Neuroendocrinology* **78** 81–89. (doi:10.1159/000071963)

Cardenas R, Lin X, Canosa LF, Luna M, Aramburo C & Peter RE 2003 Estradiol reduces pituitary responsiveness to somatostatin (SRIF-14) and down-regulates the expression of somatostatin sst2 receptors in female goldfish pituitary. *General and Comparative Endocrinology* **132** 119–124. (doi:10.1016/S0016-6480(03)00055-8)

Cerovac V, Monteserin-Garcia J, Rubinfeld H, Buchfelder M, Losa M, Florio T, Paez-Pereda M, Stalla GK & Theodoropoulou M 2010 The somatostatin analogue octreotide confers sensitivity to rapamycin treatment on pituitary tumor cells. *Cancer Research* **70** 666–674. (doi:10.1158/0008-5472.CAN-09-2951)

Cervia D 2011 The beta isoenzyme of Ca(2+)/calmodulin-dependent kinase type II as possible mediator of somatostatin functions in pituitary tumour cells. *General Physiology and Biophysics* **30** 251–262. (doi:10.4149/gpb_2011_03_251)

Cervia D & Bagnoli P 2007 An update on somatostatin receptor signaling in native systems and new insights on their pathophysiology. *Pharmacology & Therapeutics* **116** 322–341. (doi:10.1016/j.pharmthera.2007.06.010)

Cervia D, Petrucci C, Bluet-Pajot MT, Epelbaum J & Bagnoli P 2002*a* Inhibitory control of growth hormone secretion by somatostatin in rat pituitary GC cells: sst(2) but not sst(1) receptors are coupled to inhibition of single-cell intracellular free calcium concentrations. *Neuroendocrinology* **76** 99–110. (doi:10.1159/000064424)

Cervia D, Fiorini S, Pavan B, Biondi C & Bagnoli P 2002*b* Somatostatin (SRIF) modulates distinct signaling pathways in rat pituitary tumor cells; negative coupling of SRIF receptor subtypes 1 and 2 to arachidonic acid release. *Naunyn-Schmiedeberg's Archives of Pharmacology* **365** 200–209. (doi:10.1007/s00210-001-0509-7)

Cervia D, Zizzari P, Pavan B, Schuepbach E, Langenegger D, Hoyer D, Biondi C, Epelbaum J & Bagnoli P 2003 Biological activity of somatostatin receptors in GC rat tumour somatotrophs: evidence with sst1–sst5 receptor-selective nonpeptidyl agonists. *Neuropharmacology* **44** 672–685. (doi:10.1016/S0028-3908(03)00031-5)

Chen C 1997 G(o)2 and Gi3 proteins mediate the action of somatostatin on membrane Ca^{2+} and K^{+} currents in ovine pituitary somatotrophs. *Clinical and Experimental Pharmacology & Physiology* **24** 639–645. (doi:10.1111/j.1440-1681.1997.tb02105.x)

Chen C, Zhang J, Vincent JD & Israel JM 1990 Somatostatin increases voltage-dependent potassium currents in rat somatotrophs. *American Journal of Physiology* **259** C854–C861.

Colao A, Pivonello R, Di Somma C, Savastano S, Grasso LF & Lombardi G 2009 Medical therapy of pituitary adenomas: effects on tumor shrinkage. *Reviews in Endocrine & Metabolic Disorders* **10** 111–123. (doi:10.1007/s11154-008-9107-z)

Colao A, Petersenn S, Newell-Price J, Findling JW, Gu F, Maldonado M, Schoenherr U, Mills D, Salgado LR, Biller BM *et al.* 2012 A 12-month phase 3 study of pasireotide in Cushing's disease. *New England Journal of Medicine* **366** 914–924. (doi:10.1056/NEJMoa1105743)

Colao A, Bronstein M, Freda P, Gu F, Shen CC, Gadelha M, Fleseriu M, van der Lely A, Farrall A, Hermosillo Resendiz K *et al.* 2014 Pasireotide versus octreotide in acromegaly: a head-to-head superiority study. *Journal of Clinical Endocrinology and Metabolism*.

Cordoba-Chacon J, Gahete MD, Pozo-Salas AI, Martinez-Fuentes AJ, de Lecea L, Gracia-Navarro F, Kineman RD, Castano JP & Luque RM 2011 Cortistatin is not a somatostatin analogue but stimulates prolactin release and inhibits GH and ACTH in a gender-dependent fashion: potential role of ghrelin. *Endocrinology* **152** 4800–4812. (doi:10.1210/en.2011-1542)

Cordoba-Chacon J, Gahete MD, Culler MD, Castano JP, Kineman RD & Luque RM 2012 Somatostatin dramatically stimulates growth hormone release from primate somatotrophs acting at low doses via somatostatin receptor 5 and cyclic AMP. *Journal of Neuroendocrinology* **24** 453–463. (doi:10.1111/j.1365-2826.2011.02261.x)

Davis JR, Wilson EM, Vidal ME, Johnson AP, Lynch SS & Sheppard MC 1989 Regulation of growth hormone secretion and messenger ribonucleic acid accumulation in human somatotropinoma cells *in vitro*. *Journal of Clinical Endocrinology and Metabolism* **69** 704–708. (doi:10.1210/jcem-69-4-704)

Degtiar VE, Harhammer R & Nurnberg B 1997 Receptors couple to L-type calcium channels via distinct Go proteins in rat neuroendocrine cell

http://jme.endocrinology-journals.org
DOI: 10.1530/JME-14-0034

Published by Bioscientifica Ltd.

lines. *Journal of Physiology* **502** 321–333. (doi:10.1111/j.1469-7793.1997.321bk.x)

Djordjijevic D, Zhang J, Priam M, Viollet C, Gourdji D, Kordon C & Epelbaum J 1998 Effect of 17β-estradiol on somatostatin receptor expression and inhibitory effects on growth hormone and prolactin release in rat pituitary cell cultures. *Endocrinology* **139** 2272–2277. (doi:10.1210/endo.139.5.5990)

Donoghue DJ & Scanes CG 1991 Possible involvement of adenylyl cyclase–cAMP–protein kinase a pathway in somatostatin inhibition of growth hormone release from chicken pituitary cells. *General and Comparative Endocrinology* **81** 113–119. (doi:10.1016/0016-6480(91)90131-O)

Draznin B, Dahl R, Sherman N, Sussman KE & Staehelin LA 1988 Exocytosis in normal anterior pituitary cells. Quantitative correlation between growth hormone release and the morphological features of exocytosis. *Journal of Clinical Investigation* **81** 1042–1050. (doi:10.1172/JCI113415)

Drouin J 2011 Pituitary development. In *Pituitary*, 3rd edn, pp 3–19. Ed S Melmed. San Diego, California: Elsevier.

Eigler T, Ben-Shlomo A, Zhou C, Khalafi R, Ren SG & Melmed S 2014 Constitutive somatostatin receptor subtype-3 signaling suppresses growth hormone synthesis. *Molecular Endocrinology* **28** 554–564. (doi:10.1210/me.2013-1327)

Elango A, Shepherd B & Chen TT 2006 Effects of endocrine disrupters on the expression of growth hormone and prolactin mRNA in the rainbow trout pituitary. *General and Comparative Endocrinology* **145** 116–127. (doi:10.1016/j.ygcen.2005.08.003)

Estupina C, Belmar J, Tapia-Arancibia L, Astier H & Arancibia S 1997 Rapid and opposite effects of dexamethasone on *in vivo* and *in vitro* hypothalamic somatostatin release. *Experimental Brain Research* **113** 337–342. (doi:10.1007/BF02450331)

Farhy LS & Veldhuis JD 2004 Putative GH pulse renewal: periventricular somatostatinergic control of an arcuate-nuclear somatostatin and GH-releasing hormone oscillator. *American Journal of Physiology. Regulatory, Integrative and Comparative Physiology* **286** R1030–R1042. (doi:10.1152/ajpregu.00473.2003)

Fehm HL, Voigt KH, Lang R, Beinert KE, Raptis S & Pfeiffer EF 1976 Somatostatin: a potent inhibitor of ACTH-hypersecretion in adrenal insufficiency. *Klinische Wochenschrift* **54** 173–175. (doi:10.1007/BF01468882)

Fernandez G, Sanchez-Franco F, de los Frailes MT, Tolon RM, Lorenzo MJ, Lopez J & Cacicedo L 1992 Regulation of somatostatin and growth hormone-releasing factor by gonadal steroids in fetal rat hypothalamic cells in culture. *Regulatory Peptides* **42** 135–144. (doi:10.1016/0167-0115(92)90093-A)

Ferrante E, Pellegrini C, Bondioni S, Peverelli E, Locatelli M, Gelmini P, Luciani P, Peri A, Mantovani G, Bosari S *et al.* 2006 Octreotide promotes apoptosis in human somatotroph tumor cells by activating somatostatin receptor type 2. *Endocrine-Related Cancer* **13** 955–962. (doi:10.1677/erc.1.01191)

Florio T 2008 Somatostatin/somatostatin receptor signalling: phosphotyrosine phosphatases. *Molecular and Cellular Endocrinology* **286** 40–48. (doi:10.1016/j.mce.2007.08.012)

Florio T, Thellung S, Arena S, Corsaro A, Spaziante R, Gussoni G, Acuto G, Giusti M, Giordano G & Schettini G 1999 Somatostatin and its analog lanreotide inhibit the proliferation of dispersed human non-functioning pituitary adenoma cells *in vitro*. *European Journal of Endocrinology* **141** 396–408. (doi:10.1530/eje.0.1410396)

Florio T, Thellung S, Corsaro A, Bocca L, Arena S, Pattarozzi A, Villa V, Massa A, Diana F, Schettini D *et al.* 2003 Characterization of the intracellular mechanisms mediating somatostatin and lanreotide inhibition of DNA synthesis and growth hormone release from dispersed human GH-secreting pituitary adenoma cells *in vitro*. *Clinical Endocrinology* **59** 115–128. (doi:10.1046/j.1365-2265.2003.01811.x)

Foster GA & Johansson O 1985 Ultrastructural morphometric analysis of somatostatin-like immunoreactive neurones in the rat central nervous system after labelling with colloidal gold. *Brain Research* **342** 117–127. (doi:10.1016/0006-8993(85)91359-9)

Fusco A, Gunz G, Jaquet P, Dufour H, Germanetti AL, Culler MD, Barlier A & Saveanu A 2008 Somatostatinergic ligands in dopamine-sensitive and -resistant prolactinomas. *European Journal of Endocrinology* **158** 595–603. (doi:10.1530/EJE-07-0806)

Galanopoulou AS, Seidah NG & Patel YC 1995 Heterologous processing of rat prosomatostatin to somatostatin-14 by PC2: requirement for secretory cell but not the secretion granule. *Biochemical Journal* **311** 111–118.

Galli G, Zonefrati R, Gozzini A, Mavilia C, Martineti V, Tognarini I, Nesi G, Marcucci T, Tonelli F, Tommasi M *et al.* 2006 Characterization of the functional and growth properties of long-term cell cultures established from a human somatostatinoma. *Endocrine-Related Cancer* **13** 79–93. (doi:10.1677/erc.1.00988)

Gancel A, Vuillermet P, Legrand A, Catus F, Thomas F & Kuhn JM 1994 Effects of a slow-release formulation of the new somatostatin analogue lanreotide in TSH-secreting pituitary adenomas. *Clinical Endocrinology* **40** 421–428. (doi:10.1111/j.1365-2265.1994.tb03941.x)

Giustina A & Veldhuis JD 1998 Pathophysiology of the neuroregulation of growth hormone secretion in experimental animals and the human. *Endocrine Reviews* **19** 717–797. (doi:10.1210/edrv.19.6.0353)

Gooren LJ, Harmsen-Louman W & van Kessel H 1984 Somatostatin inhibits prolactin release from the lactotroph primed with oestrogen and cyproterone acetate in man. *Journal of Endocrinology* **103** 333–335. (doi:10.1677/joe.0.1030333)

Goth MI, Lyons CE Jr, Ellwood MR, Barrett JR & Thorner MO 1996 Chronic estrogen treatment in male rats reveals mammosomatotropes and allows inhibition of prolactin secretion by somatostatin. *Endocrinology* **137** 274–280. (doi:10.1210/endo.137.1.8536623)

Grant M, Alturaihi H, Jaquet P, Collier B & Kumar U 2008 Cell growth inhibition and functioning of human somatostatin receptor type 2 are modulated by receptor heterodimerization. *Molecular Endocrinology* **22** 2278–2292. (doi:10.1210/me.2007-0334)

Greenman Y & Stern N 2009 Non-functioning pituitary adenomas. *Best Practice & Research. Clinical Endocrinology & Metabolism* **23** 625–638. (doi:10.1016/j.beem.2009.05.005)

Gruszka A, Ren SG, Dong J, Culler MD & Melmed S 2007 Regulation of growth hormone and prolactin gene expression and secretion by chimeric somatostatin–dopamine molecules. *Endocrinology* **148** 6107–6114. (doi:10.1210/en.2007-0378)

Gruszka A, Culler MD & Melmed S 2012 Somatostatin analogs and chimeric somatostatin–dopamine molecules differentially regulate human growth hormone and prolactin gene expression and secretion *in vitro*. *Molecular and Cellular Endocrinology* **362** 104–109. (doi:10.1016/j.mce.2012.05.020)

Guillermet-Guibert J, Saint-Laurent N, Davenne L, Rochaix P, Cuvillier O, Culler MD, Pradayrol L, Buscail L, Susini C & Bousquet C 2007 Novel synergistic mechanism for sst2 somatostatin and TNFα receptors to induce apoptosis: crosstalk between NF-κB and JNK pathways. *Cell Death and Differentiation* **14** 197–208. (doi:10.1038/sj.cdd.4401939)

Hao L, Li M, Dai J, Wu Q, Liu Y, Liu S & Zhang Y 2010 Reduced somatostatin in hypothalamus of young male mouse increases local but not circulatory GH. *Neurochemical Research* **35** 143–151. (doi:10.1007/s11064-009-0039-0)

Hassan HA, Enright WJ, Tucker HA & Merkel RA 2001 Estrogen and androgen elicit growth hormone release via dissimilar patterns of hypothalamic neuropeptide secretion. *Steroids* **66** 71–80. (doi:10.1016/S0039-128X(00)00168-9)

Heisler S, Reisine TD, Hook VY & Axelrod J 1982 Somatostatin inhibits multireceptor stimulation of cyclic AMP formation and corticotropin secretion in mouse pituitary tumor cells. *PNAS* **79** 6502–6506. (doi:10.1073/pnas.79.21.6502)

Hipkin RW, Friedman J, Clark RB, Eppler CM & Schonbrunn A 1997 Agonist-induced desensitization, internalization, and phosphorylation of the sst2A somatostatin receptor. *Journal of Biological Chemistry* **272** 13869–13876. (doi:10.1074/jbc.272.21.13869)

http://jme.endocrinology-journals.org
DOI: 10.1530/JME-14-0034

Published by Bioscientifica Ltd.

van der Hoek J, Waaijers M, van Koetsveld PM, Sprij-Mooij D, Feelders RA, Schmid HA, Schoeffter P, Hoyer D, Cervia D, Taylor JE *et al.* 2005 Distinct functional properties of native somatostatin receptor subtype 5 compared with subtype 2 in the regulation of ACTH release by corticotroph tumor cells. *American Journal of Physiology. Endocrinology and Metabolism* **289** (Suppl 1) E278–E287. (doi:10.1152/ajpendo.00004.2005)

van der Hoek J, Lamberts SW & Hofland LJ 2007 Preclinical and clinical experiences with the role of somatostatin receptors in the treatment of pituitary adenomas. *European Journal of Endocrinology* **156** S45–S51. (doi:10.1530/eje.1.02350)

Hofland LJ 2008 Somatostatin and somatostatin receptors in Cushing's disease. *Molecular and Cellular Endocrinology* **286** 199–205. (doi:10.1016/j.mce.2007.10.015)

Hofland LJ, van der Hoek J, van Koetsveld PM, de Herder WW, Waaijers M, Sprij-Mooij D, Bruns C, Weckbecker G, Feelders R, van der Lely AJ *et al.* 2004 The novel somatostatin analog SOM230 is a potent inhibitor of hormone release by growth hormone- and prolactin-secreting pituitary adenomas *in vitro*. *Journal of Clinical Endocrinology and Metabolism* **89** 1577–1585. (doi:10.1210/jc.2003-031344)

Holl RW, Thorner MO, Mandell GL, Sullivan JA, Sinha YN & Leong DA 1988 Spontaneous oscillations of intracellular calcium and growth hormone secretion. *Journal of Biological Chemistry* **263** 9682–9685.

Honegger J, Spagnoli A, D'Urso R, Navarra P, Tsagarakis S, Besser GM & Grossman AB 1991 Interleukin-1 beta modulates the acute release of growth hormone-releasing hormone and somatostatin from rat hypothalamus *in vitro*, whereas tumor necrosis factor and interleukin-6 have no effect. *Endocrinology* **129** 1275–1282. (doi:10.1210/endo-129-3-1275)

Horiguchi K, Yamada M, Satoh T, Hashimoto K, Hirato J, Tosaka M, Yamada S & Mori M 2009 Transcriptional activation of the mixed lineage leukemia–p27Kip1 pathway by a somatostatin analogue. *Clinical Cancer Research* **15** 2620–2629. (doi:10.1158/1078-0432.CCR-08-2473)

Hubina E, Nanzer AM, Hanson MR, Ciccarelli E, Losa M, Gaia D, Papotti M, Terreni MR, Khalaf S, Jordan S *et al.* 2006 Somatostatin analogues stimulate p27 expression and inhibit the MAP kinase pathway in pituitary tumours. *European Journal of Endocrinology* **155** 371–379. (doi:10.1530/eje.1.02213)

Ikuyama S, Nawata H, Kato K, Natori S & Ibayashi H 1987 Phorbol ester and phospholipase C-induced growth hormone secretion from pituitary somatotroph adenoma cells in culture: effects of somatostatin, bromocriptine, and pertussis toxin. *Journal of Clinical Endocrinology and Metabolism* **64** 572–577. (doi:10.1210/jcem-64-3-572)

Iwasaki K, Fujii A, Merchenthaler I, Groot K & Arimura A 1987 The stimulation of somatostatin release by hpGRF44 from rat hypothalamic cells and fragments *in vitro*. *Tokai Journal of Experimental and Clinical Medicine* **12** 117–124.

James RA, Sarapura VD, Bruns C, Raulf F, Dowding JM, Gordon DF, Wood WM & Ridgway EC 1997 Thyroid hormone-induced expression of specific somatostatin receptor subtypes correlates with involution of the TtT-97 murine thyrotrope tumor. *Endocrinology* **138** 719–724. (doi:10.1210/endo.138.2.4951)

Jaquet P, Ouafik L, Saveanu A, Gunz G, Fina F, Dufour H, Culler MD, Moreau JP & Enjalbert A 1999 Quantitative and functional expression of somatostatin receptor subtypes in human prolactinomas. *Journal of Clinical Endocrinology and Metabolism* **84** 3268–3276. (doi:10.1210/jcem.84.9.5962)

Judd AM, Koike K & MacLeod RM 1986 A possible role of arachidonate metabolism in the mechanism of prolactin release. *American Journal of Physiology* **250** E288–E295.

Kajimura S, Hirano T, Visitacion N, Moriyama S, Aida K & Grau EG 2003 Dual mode of cortisol action on GH/IGF-I/IGF binding proteins in the tilapia, *Oreochromis mossambicus*. *Journal of Endocrinology* **178** 91–99. (doi:10.1677/joe.0.1780091)

Karanth S, Aguila MC & McCann SM 1993 The influence of interleukin-2 on the release of somatostatin and growth hormone-releasing hormone by mediobasal hypothalamus. *Neuroendocrinology* **58** 185–190.

Kawano H & Daikoku S 1988 Somatostatin-containing neuron systems in the rat hypothalamus: retrograde tracing and immunohistochemical studies. *Journal of Comparative Neurology* **271** 293–299. (doi:10.1002/cne.902710209)

Khattak MN, Buchfelder M, Kleindienst A, Schofl C & Kremenevskaja N 2010 CRH and SRIF have opposite effects on the Wnt/β-catenin signalling pathway through PKA/GSK-3β in corticotroph pituitary cells. *Cancer Investigation* **28** 797–805. (doi:10.3109/07357907.2010.494318)

Kimura N, Tomizawa S & Arai KN 1998 Chronic treatment with estrogen up-regulates expression of sst2 messenger ribonucleic acid (mRNA) but down-regulates expression of sst5 mRNA in rat pituitaries. *Endocrinology* **139** 1573–1580. (doi:10.1210/endo.139.4.5952)

Kleuss C, Scherubl H, Hescheler J, Schultz G & Wittig B 1992 Different beta-subunits determine G-protein interaction with transmembrane receptors. *Nature* **358** 424–426. (doi:10.1038/358424a0)

Kleuss C, Scherubl H, Hescheler J, Schultz G & Wittig B 1993 Selectivity in signal transduction determined by gamma subunits of heterotrimeric G proteins. *Science* **259** 832–834. (doi:10.1126/science.8094261)

Klibanski A, Alexander JM, Bikkal HA, Hsu DW, Swearingen B & Zervas NT 1991 Somatostatin regulation of glycoprotein hormone and free subunit secretion in clinically nonfunctioning and somatotroph adenomas *in vitro*. *Journal of Clinical Endocrinology and Metabolism* **73** 1248–1255. (doi:10.1210/jcem-73-6-1248)

Koch BD, Dorflinger LJ & Schonbrunn A 1985 Pertussis toxin blocks both cyclic AMP-mediated and cyclic AMP-independent actions of somatostatin. Evidence for coupling of Ni to decreases in intracellular free calcium. *Journal of Biological Chemistry* **260** 13138–13145.

Korbonits M, Little JA, Forsling ML, Tringali G, Costa A, Navarra P, Trainer PJ & Grossman AB 1999 The effect of growth hormone secretagogues and neuropeptide Y on hypothalamic hormone release from acute rat hypothalamic explants. *Journal of Neuroendocrinology* **11** 521–528. (doi:10.1046/j.1365-2826.1999.00353.x)

Kraicer J & Spence JW 1981 Release of growth hormone from purified somatotrophs: use of high K^+ and the ionophore A23187 to elucidate interrelations among Ca^{++}, adenosine 3′,5′-monophosphate, and somatostatin. *Endocrinology* **108** 651–657. (doi:10.1210/endo-108-2-651)

Kraicer J, Gajewski TC & Moor BC 1985 Release of pro-opiomelanocortin-derived peptides from the pars intermedia and pars distalis of the rat pituitary: effect of corticotrophin-releasing factor and somatostatin. *Neuroendocrinology* **41** 363–373. (doi:10.1159/000124203)

Lamberts SW, Zuyderwijk J, den Holder F, van Koetsveld P & Hofland L 1989*a* Studies on the conditions determining the inhibitory effect of somatostatin on adrenocorticotropin, prolactin and thyrotropin release by cultured rat pituitary cells. *Neuroendocrinology* **50** 44–50. (doi:10.1159/000125200)

Lamberts SW, Uitterlinden P & Klijn JM 1989*b* The effect of the long-acting somatostatin analogue SMS 201-995 on ACTH secretion in Nelson's syndrome and Cushing's disease. *Acta Endocrinologica* **120** 760–766.

de Lecea L & Castano JP 2006 Cortistatin: not just another somatostatin analog. *Nature Clinical Practice. Endocrinology & Metabolism* **2** 356–357. (doi:10.1038/ncpendmet0219)

Lee SC & Shin SH 1996 Somatostatin does not inhibit prolactin synthesis in normal male rat pituitary cells but inhibits prolactin synthesis in estradiol-primed pituitary cells. *Journal of Endocrinology* **148** 69–76. (doi:10.1677/joe.0.1480069)

Lengyel AM, Grossman A, Bouloux PM, Rees LH & Besser GM 1985 Effects of dopamine and morphine on immunoreactive somatostatin and LH-releasing hormone secretion from hypothalamic fragments *in vitro*. *Journal of Endocrinology* **106** 317–322. (doi:10.1677/joe.0.1060317)

Liu YF, Jakobs KH, Rasenick MM & Albert PR 1994 G protein specificity in receptor–effector coupling. Analysis of the roles of G0 and Gi2 in GH4C1 pituitary cells. *Journal of Biological Chemistry* **269** 13880–13886.

http://jme.endocrinology-journals.org
DOI: 10.1530/JME-14-0034

Published by Bioscientifica Ltd.

Liu HS, Hu ZT, Zhou KM, Jiu YM, Yang H, Wu ZX & Xu T 2006 Heterogeneity of the Ca^{2+} sensitivity of secretion in a pituitary gonadotrope cell line and its modulation by protein kinase C and Ca^{2+}. *Journal of Cellular Physiology* **207** 668–674. (doi:10.1002/jcp.20598)

Liu Q, Bee MS & Schonbrunn A 2009 Site specificity of agonist and second messenger-activated kinases for somatostatin receptor subtype 2A (Sst2A) phosphorylation. *Molecular Pharmacology* **76** 68–80. (doi:10.1124/mol.108.054262)

Lohrer P, Gloddek J, Hopfner U, Losa M, Uhl E, Pagotto U, Stalla GK & Renner U 2001 Vascular endothelial growth factor production and regulation in rodent and human pituitary tumor cells *in vitro*. *Neuroendocrinology* **74** 95–105. (doi:10.1159/000054675)

Low MJ, Otero-Corchon V, Parlow AF, Ramirez JL, Kumar U, Patel YC & Rubinstein M 2001 Somatostatin is required for masculinization of growth hormone-regulated hepatic gene expression but not of somatic growth. *Journal of Clinical Investigation* **107** 1571–1580. (doi:10.1172/JCI11941)

Luque RM, Park S, Peng XD, Delgado E, Gracia-Navarro F, Kineman RD, Malagon MM & Castano JP 2004 Homologous and heterologous *in vitro* regulation of pig pituitary somatostatin receptor subtypes, sst1, sst2 and sst5 mRNA. *Journal of Molecular Endocrinology* **32** 437–448. (doi:10.1677/jme.0.0320437)

Luque RM, Rodriguez-Pacheco F, Tena-Sempere M, Gracia-Navarro F, Malagon MM & Castano JP 2005 Differential contribution of nitric oxide and cGMP to the stimulatory effects of growth hormone-releasing hormone and low-concentration somatostatin on growth hormone release from somatotrophs. *Journal of Neuroendocrinology* **17** 577–582. (doi:10.1111/j.1365-2826.2005.01345.x)

Luque RM, Duran-Prado M, Garcia-Navarro S, Gracia-Navarro F, Kineman RD, Malagon MM & Castano JP 2006*a* Identification of the somatostatin receptor subtypes (sst) mediating the divergent, stimulatory/inhibitory actions of somatostatin on growth hormone secretion. *Endocrinology* **147** 2902–2908. (doi:10.1210/en.2005-1559)

Luque RM, Gahete MD, Hochgeschwender U & Kineman RD 2006*b* Evidence that endogenous SST inhibits ACTH and ghrelin expression by independent pathways. *American Journal of Physiology. Endocrinology and Metabolism* **291** E395–E403. (doi:10.1152/ajpendo.00038.2006)

Maeda K & Frohman LA 1980 Release of somatostatin and thyrotropin-releasing hormone from rat hypothalamic fragments *in vitro*. *Endocrinology* **106** 1837–1842. (doi:10.1210/endo-106-6-1837)

Matsuno A, Itoh J, Takekoshi S, Itoh Y, Ohsugi Y, Katayama H, Nagashima T & Osamura RY 2003 Dynamics of subcellular organelles, growth hormone, Rab3B, SNAP-25, and syntaxin in rat pituitary cells caused by growth hormone releasing hormone and somatostatin. *Microscopy Research and Technique* **62** 232–239. (doi:10.1002/jemt.10364)

Melmed S 2006 Medical progress: acromegaly. *New England Journal of Medicine* **355** 2558–2573. (doi:10.1056/NEJMra062453)

Millar RP, Klaff LJ, Barron J, Levitt NS & Ling N 1982 Somatostatin-28 inhibits LHRH-stimulated gonadotrophin secretion in man. *Clinical Endocrinology* **17** 103–107. (doi:10.1111/j.1365-2265.1982.tb01569.x)

Morishita M, Iwasaki Y, Onishi A, Asai M, Mutsuga N, Yoshida M, Oiso Y, Inoue K & Murohara T 2003 The effects of GH-releasing hormone/somatostatin on the 5′-promoter activity of the GH gene *in vitro*. *Journal of Molecular Endocrinology* **31** 441–448. (doi:10.1677/jme.0.0310441)

Namba H, Morita S & Melmed S 1989 Insulin-like growth factor-I action on growth hormone secretion and messenger ribonucleic acid levels: interaction with somatostatin. *Endocrinology* **124** 1794–1799. (doi:10.1210/endo-124-4-1794)

Negro-Vilar A, Ojeda SR, Arimura A & McCann SM 1978 Dopamine and norepinephrine stimulate somatostatin release by median eminence fragments *in vitro*. *Life Sciences* **23** 1493–1497.

Norman M, Moldovan S, Seghers V, Wang XP, DeMayo FJ & Brunicardi FC 2002 Sulfonylurea receptor knockout causes glucose intolerance in mice that is not alleviated by concomitant somatostatin subtype receptor 5 knockout. *Annals of Surgery* **235** 767–774. (doi:10.1097/00000658-200206000-00003)

Oberg K & Eriksson B 2005 Endocrine tumours of the pancreas. *Best Practice & Research. Clinical Gastroenterology* **19** 753–781. (doi:10.1016/j.bpg.2005.06.002)

Otsuka F, Tsukamoto N, Miyoshi T, Iwasaki Y & Makino H 2012 BMP action in the pituitary: its possible role in modulating somatostatin sensitivity in pituitary tumor cells. *Molecular and Cellular Endocrinology* **349** 105–110. (doi:10.1016/j.mce.2011.10.017)

Panetta R & Patel YC 1995 Expression of mRNA for all five human somatostatin receptors (hSSTR1–5) in pituitary tumors. *Life Sciences* **56** 333–342. (doi:10.1016/0024-3205(94)00956-2)

Park C, Yang I, Woo J, Kim S, Kim J, Kim Y, Sohn S, Kim E, Lee M, Park H *et al.* 2004 Somatostatin (SRIF) receptor subtype 2 and 5 gene expression in growth hormone-secreting pituitary adenomas: the relationship with endogenous srif activity and response to octreotide. *Endocrine Journal* **51** 227–236. (doi:10.1507/endocrj.51.227)

van der Pas R, Feelders RA, Gatto F, de Bruin C, Pereira AM, van Koetsveld PM, Sprij-Mooij DM, Waaijers AM, Dogan F, Schulz S *et al.* 2013 Preoperative normalization of cortisol levels in Cushing's disease after medical treatment: consequences for somatostatin and dopamine receptor subtype expression and *in vitro* response to somatostatin analogs and dopamine agonists. *Journal of Clinical Endocrinology and Metabolism* **98** E1880–E1890. (doi:10.1210/jc.2013-1987)

Patel YC, Greenwood M, Kent G, Panetta R & Srikant CB 1993 Multiple gene transcripts of the somatostatin receptor SSTR2: tissue selective distribution and cAMP regulation. *Biochemical and Biophysical Research Communications* **192** 288–294. (doi:10.1006/bbrc.1993.1412)

Patel YC 1999 Somatostatin and its receptor family. *Frontiers in Neuroendocrinology* **20** 157–198. (doi:10.1006/frne.1999.0183)

Patel YC & Srikant CB 1986 Somatostatin mediation of adenohypophysial secretion. *Annual Review of Physiology* **48** 551–567. (doi:10.1146/annurev.ph.48.030186.003003)

Peterfreund RA & Vale WW 1983 Muscarinic cholinergic stimulation of somatostatin secretion from long term dispersed cell cultures of fetal rat hypothalamus: inhibition by gamma-aminobutyric acid and serotonin. *Endocrinology* **112** 526–534. (doi:10.1210/endo-112-2-526)

Petersenn S, Rasch AC, Presch S, Beil FU & Schulte HM 1999 Genomic structure and transcriptional regulation of the human somatostatin receptor type 2. *Molecular and Cellular Endocrinology* **157** 75–85. (doi:10.1016/S0303-7207(99)00161-6)

Petersenn S, Farrall AJ, Block C, Melmed S, Schopohl J, Caron P, Cuneo R, Kleinberg D, Colao A, Ruffin M *et al.* 2014*a* Long-term efficacy and safety of subcutaneous pasireotide in acromegaly: results from an open-ended, multicenter, phase II extension study. *Pituitary* **17** 132–140. (doi:10.1007/s11102-013-0478-0)

Petersenn S, Farrall AJ, De Block C, Melmed S, Schopohl J, Caron P, Cuneo R, Kleinberg D, Colao A, Ruffin M *et al.* 2014*b* Erratum to: Long-term efficacy and safety of subcutaneous pasireotide in acromegaly: results from an open-ended, multicenter, phase II extension study. *Pituitary* **17** 141. (doi:10.1007/s11102-013-0491-3)

Petrucci C, Cervia D, Buzzi M, Biondi C & Bagnoli P 2000 Somatostatin-induced control of cytosolic free calcium in pituitary tumour cells. *British Journal of Pharmacology* **129** 471–484. (doi:10.1038/sj.bjp.0703075)

Peverelli E, Mantovani G, Calebiro D, Doni A, Bondioni S, Lania A, Beck-Peccoz P & Spada A 2008 The third intracellular loop of the human somatostatin receptor 5 is crucial for arrestin binding and receptor internalization after somatostatin stimulation. *Molecular Endocrinology* **22** 676–688. (doi:10.1210/me.2007-0068)

Peverelli E, Busnelli M, Vitali E, Giardino E, Gales C, Lania AG, Beck-Peccoz P, Chini B, Mantovani G & Spada A 2013 Specific roles of G(i) protein family members revealed by dissecting SST5 coupling in

http://jme.endocrinology-journals.org
DOI: 10.1530/JME-14-0034

Published by Bioscientifica Ltd.

human pituitary cells. *Journal of Cell Science* **126** 638–644. (doi:10.1242/jcs.116434)

Pfeiffer M, Koch T, Schroder H, Laugsch M, Hollt V & Schulz S 2002 Heterodimerization of somatostatin and opioid receptors cross-modulates phosphorylation, internalization, and desensitization. *Journal of Biological Chemistry* **277** 19762–19772. (doi:10.1074/jbc.M110373200)

Pillon D, Caraty A, Fabre-Nys C, Lomet D, Cateau M & Bruneau G 2004 Regulation by estradiol of hypothalamic somatostatin gene expression: possible involvement of somatostatin in the control of luteinizing hormone secretion in the ewe. *Biology of Reproduction* **71** 38–44. (doi:10.1095/biolreprod.103.023689)

Poll F, Lehmann D, Illing S, Ginj M, Jacobs S, Lupp A, Stumm R & Schulz S 2010 Pasireotide and octreotide stimulate distinct patterns of sst2A somatostatin receptor phosphorylation. *Molecular Endocrinology* **24** 436–446. (doi:10.1210/me.2009-0315)

Presky DH & Schonbrunn A 1988 Iodination of [Tyr11]somatostatin yields a super high affinity ligand for somatostatin receptors in GH4C1 pituitary cells. *Molecular Pharmacology* **34** 651–658.

Puente E, Saint-Laurent N, Torrisani J, Furet C, Schally AV, Vaysse N, Buscail L & Susini C 2001 Transcriptional activation of mouse sst2 somatostatin receptor promoter by transforming growth factor-beta, Involvement of Smad4. *Journal of Biological Chemistry* **276** 13461–13468. (doi:10.1074/jbc.M010981200)

Quintela M, Senaris R, Heiman ML, Casanueva FF & Dieguez C 1997*a* Leptin inhibits *in vitro* hypothalamic somatostatin secretion and somatostatin mRNA levels. *Endocrinology* **138** 5641–5644.

Quintela M, Senaris RM & Dieguez C 1997*b* Transforming growth factor-betas inhibit somatostatin messenger ribonucleic acid levels and somatostatin secretion in hypothalamic cells in culture. *Endocrinology* **138** 4401–4409. (doi:10.1210/endo.138.10.5467)

Ramirez JL, Gracia-Navarro F, Garcia-Navarro S, Torronteras R, Malagon MM & Castano JP 2002 Somatostatin stimulates GH secretion in two porcine somatotrope subpopulations through a cAMP-dependent pathway. *Endocrinology* **143** 889–897. (doi:10.1210/endo.143.3.8685)

Reichlin S 1983 Somatostatin. *New England Journal of Medicine* **309** 1495–1501. (doi:10.1056/NEJM198312153092406)

Reisine T & Axelrod J 1983 Prolonged somatostatin pretreatment desensitizes somatostatin's inhibition of receptor-mediated release of adrenocorticotropin hormone and sensitizes adenylate cyclase. *Endocrinology* **113** 811–813. (doi:10.1210/endo-113-2-811)

Ren SG, Taylor J, Dong J, Yu R, Culler MD & Melmed S 2003 Functional association of somatostatin receptor subtypes 2 and 5 in inhibiting human growth hormone secretion. *Journal of Clinical Endocrinology and Metabolism* **88** 4239–4245. (doi:10.1210/jc.2003-030303)

Resmini E, Dadati P, Ravetti JL, Zona G, Spaziante R, Saveanu A, Jaquet P, Culler MD, Bianchi F, Rebora A *et al.* 2007 Rapid pituitary tumor shrinkage with dissociation between antiproliferative and antisecretory effects of a long-acting octreotide in an acromegalic patient. *Journal of Clinical Endocrinology and Metabolism* **92** 1592–1599. (doi:10.1210/jc.2006-2084)

Richardson SB, Hollander CS, D'Eletto R, Greenleaf PW & Thaw C 1980 Acetylcholine inhibits the release of somatostatin from rat hypothalamus *in vitro*. *Endocrinology* **107** 122–129. (doi:10.1210/endo-107-1-122)

Richardson SB, Hollander CS, Prasad JA & Hirooka Y 1981 Somatostatin release from rat hypothalamus *in vitro*: effects of melatonin and serotonin. *Endocrinology* **109** 602–606. (doi:10.1210/endo-109-2-602)

Richardson S, Twente S & Audhya T 1988 GHRF causes biphasic stimulation of SRIF secretion from rat hypothalamic cells. *American Journal of Physiology* **255** E829–E832.

Richardson SB & Twente S 1990 Inhibition of hypothalamic somatostatin release by beta-adrenergic antagonists. *Endocrinology* **126** 1043–1046. (doi:10.1210/endo-126-2-1043)

Roosterman D, Kempkes C, Cottrell GS, Padilla BE, Bunnett NW, Turck CW & Steinhoff M 2008 Endothelin-converting enzyme-1 degrades internalized somatostatin-14. *Endocrinology* **149** 2200–2207. (doi:10.1210/en.2007-1628)

Samuels MH, Henry P & Ridgway EC 1992 Effects of dopamine and somatostatin on pulsatile pituitary glycoprotein secretion. *Journal of Clinical Endocrinology and Metabolism* **74** 217–222. (doi:10.1210/jcem.74.1.1345783)

Sarret P, Nouel D, Dal Farra C, Vincent JP, Beaudet A & Mazella J 1999 Receptor-mediated internalization is critical for the inhibition of the expression of growth hormone by somatostatin in the pituitary cell line AtT-20. *Journal of Biological Chemistry* **274** 19294–19300. (doi:10.1074/jbc.274.27.19294)

Sassolas G, Khalfallah Y, Chayvialle JA, Cohen R, Merabet S, Casez JP, Calvet P & Cabrera P 1989 Effects of the somatostatin analog BIM 23014 on the secretion of growth hormone, thyrotropin, and digestive peptides in normal men. *Journal of Clinical Endocrinology and Metabolism* **68** 239–246. (doi:10.1210/jcem-68-2-239)

Scarborough DE, Lee SL, Dinarello CA & Reichlin S 1989 Interleukin-1 beta stimulates somatostatin biosynthesis in primary cultures of fetal rat brain. *Endocrinology* **124** 549–551. (doi:10.1210/endo-124-1-549)

Schussler N, Farnoud R, Rauch C, Roche M, Berthet M, Thomas F, Peillon F & Bayet MC 1994 Effect of the slow-release formulation of somatuline (BIM 23014) on estrogen-induced hyperprolactinemia and lactotroph hyperplasia in the female rat. *Neuropeptides* **26** 399–404. (doi:10.1016/0143-4179(94)90025-6)

Seifert R & Wenzel-Seifert K 2002 Constitutive activity of G-protein-coupled receptors: cause of disease and common property of wild-type receptors. *Naunyn-Schmiedeberg's Archives of Pharmacology* **366** 381–416. (doi:10.1007/s00210-002-0588-0)

Senaris RM, Lago F, Lewis MD, Dominguez F, Scanlon MF & Dieguez C 1992 Differential effects of *in vivo* estrogen administration on hypothalamic growth hormone releasing hormone and somatostatin gene expression. *Neuroscience Letters* **141** 123–126. (doi:10.1016/0304-3940(92)90349-C)

Sheppard MC, Kronheim S & Pimstone BL 1978 Stimulation by growth hormone of somatostatin release from the rat hypothalamus *in vitro*. *Clinical Endocrinology* **9** 583–586. (doi:10.1111/j.1365-2265.1978.tb01518.x)

Sheppard MC, Kronheim S & Pimstone BL 1979 Effect of substance P, neurotensin and the enkephalins on somatostatin release from the rat hypothalamus *in vitro*. *Journal of Neurochemistry* **32** 647–649. (doi:10.1111/j.1471-4159.1979.tb00400.x)

Shimada O, Tosaka-Shimada H & Ishikawa H 1990 Morphological effects of somatostatin on rat somatotrophs previously activated by growth hormone-releasing factor. *Cell and Tissue Research* **261** 219–229. (doi:10.1007/BF00318663)

Shimatsu A, Kato Y, Matsushita N, Katakami H, Yanaihara N & Imura H 1982 Effects of glucagon, Neurotensin, and vasoactive intestinal polypeptide on somatostatin release from perifused rat hypothalamus. *Endocrinology* **110** 2113–2117. (doi:10.1210/endo-110-6-2113)

Shimon I, Yan X, Taylor JE, Weiss MH, Culler MD & Melmed S 1997*a* Somatostatin receptor (SSTR) subtype-selective analogues differentially suppress *in vitro* growth hormone and prolactin in human pituitary adenomas, Novel potential therapy for functional pituitary tumors. *Journal of Clinical Investigation* **100** 2386–2392. (doi:10.1172/JCI119779)

Shimon I, Taylor JE, Dong JZ, Bitonte RA, Kim S, Morgan B, Coy DH, Culler MD & Melmed S 1997*b* Somatostatin receptor subtype specificity in human fetal pituitary cultures, Differential role of SSTR2 and SSTR5 for growth hormone, thyroid-stimulating hormone, and prolactin regulation. *Journal of Clinical Investigation* **99** 789–798. (doi:10.1172/JCI119225)

Simard J, Labrie F & Gossard F 1986 Regulation of growth hormone mRNA and pro-opiomelanocortin mRNA levels by cyclic AMP in rat anterior pituitary cells in culture. *DNA* **5** 263–270. (doi:10.1089/dna.1986.5.263)

http://jme.endocrinology-journals.org
DOI: 10.1530/JME-14-0034

Published by Bioscientifica Ltd.

Simonian SX, Murray HE, Gillies GE & Herbison AE 1998 Estrogen-dependent ontogeny of sex differences in somatostatin neurons of the hypothalamic periventricular nucleus. *Endocrinology* **139** 1420–1428. (doi:10.1210/endo.139.3.5814)

Soya H & Suzuki M 1990 A possible role of hypothalamic somatostatin in the maintenance of rat pituitary responsiveness to growth hormone-releasing factor. *Endocrinology* **126** 285–291. (doi:10.1210/endo-126-1-285)

Spada A, Reza-Elahi F, Lania A, Bassetti M & Atti E 1990 Inhibition of basal and corticotropin-releasing hormone-stimulated adenylate cyclase activity and cytosolic Ca^{2+} levels by somatostatin in human corticotropin-secreting pituitary adenomas. *Journal of Clinical Endocrinology and Metabolism* **70** 1262–1268. (doi:10.1210/jcem-70-5-1262)

Spier AD & de Lecea L 2000 Cortistatin: a member of the somatostatin neuropeptide family with distinct physiological functions. *Brain Research. Brain Research Reviews* **33** 228–241. (doi:10.1016/S0165-0173(00)00031-X)

Spoudeas HA, Matthews DR, Brook CG & Hindmarsh PC 1992 The effect of changing somatostatin tone on the pituitary growth hormone and thyroid-stimulating hormone responses to their respective releasing factor stimuli. *Journal of Clinical Endocrinology and Metabolism* **75** 453–458. (doi:10.1210/jcem.75.2.1353504)

Stafford PJ, Kopelman PG, Davidson K, McLoughlin L, White A, Rees LH, Besser GM, Coy DH & Grossman A 1989 The pituitary–adrenal response to CRF-41 is unaltered by intravenous somatostatin in normal subjects. *Clinical Endocrinology* **30** 661–666. (doi:10.1111/j.1365-2265.1989.tb00272.x)

Starcevic V, Milosevic V, Brkic B & Severs WB 2002 Somatostatin affects morphology and secretion of pituitary luteinizing hormone (LH) cells in male rats. *Life Sciences* **70** 3019–3027. (doi:10.1016/S0024-3205(02)01551-5)

Strowski MZ, Kohler M, Chen HY, Trumbauer ME, Li Z, Szalkowski D, Gopal-Truter S, Fisher JK, Schaeffer JM, Blake AD *et al.* 2003 Somatostatin receptor subtype 5 regulates insulin secretion and glucose homeostasis. *Molecular Endocrinology* **17** 93–106. (doi:10.1210/me.2001-0035)

Sugihara H, Minami S, Okada K, Kamegai J, Hasegawa O & Wakabayashi I 1993 Somatostatin reduces transcription of the growth hormone gene in rats. *Endocrinology* **132** 1225–1229. (doi:10.1210/endo.132.3.7679974)

Taboada GF, Luque RM, Neto LV, Machado Ede O, Sbaffi BC, Domingues RC, Marcondes JB, Chimelli LM, Fontes R, Niemeyer P *et al.* 2008 Quantitative analysis of somatostatin receptor subtypes (1–5) gene expression levels in somatotropinomas and correlation to *in vivo* hormonal and tumor volume responses to treatment with octreotide LAR. *European Journal of Endocrinology* **158** 295–303. (doi:10.1530/EJE-07-0562)

Takano K, Yasufuku-Takano J, Kozasa T, Nakajima S & Nakajima Y 1997 Different G proteins mediate somatostatin-induced inward rectifier K^+ currents in murine brain and endocrine cells. *Journal of Physiology* **502** 559–567. (doi:10.1111/j.1469-7793.1997.559bj.x)

Tallent M & Reisine T 1992 Giα1 selectively couples somatostatin receptors to adenylyl cyclase in pituitary-derived AtT-20 cells. *Molecular Pharmacology* **41** 452–455.

Tanner JW, Davis SK, McArthur NH, French JT & Welsh TH Jr 1990 Modulation of growth hormone (GH) secretion and GH mRNA levels by GH-releasing factor, somatostatin and secretagogues in cultured bovine adenohypophysial cells. *Journal of Endocrinology* **125** 109–115. (doi:10.1677/joe.0.1250109)

Tentler JJ, Hadcock JR & Gutierrez-Hartmann A 1997 Somatostatin acts by inhibiting the cyclic 3′,5′-adenosine monophosphate (cAMP)/protein kinase A pathway, cAMP response element-binding protein (CREB) phosphorylation, and CREB transcription potency. *Molecular Endocrinology* **11** 859–866. (doi:10.1210/mend.11.7.9943)

Theodoropoulou M, Zhang J, Laupheimer S, Paez-Pereda M, Erneux C, Florio T, Pagotto U & Stalla GK 2006 Octreotide, a somatostatin analogue, mediates its antiproliferative action in pituitary tumor cells by altering phosphatidylinositol 3-kinase signaling and inducing Zac1 expression. *Cancer Research* **66** 1576–1582. (doi:10.1158/0008-5472.CAN-05-1189)

Theodoropoulou M, Tichomirowa MA, Sievers C, Yassouridis A, Arzberger T, Hougrand O, Deprez M, Daly AF, Petrossians P, Pagotto U *et al.* 2009 Tumor ZAC1 expression is associated with the response to somatostatin analog therapy in patients with acromegaly. *International Journal of Cancer* **125** 2122–2126. (doi:10.1002/ijc.24602)

Tsaneva-Atanasova K, Sherman A, van Goor F & Stojilkovic SS 2007 Mechanism of spontaneous and receptor-controlled electrical activity in pituitary somatotrophs: experiments and theory. *Journal of Neurophysiology* **98** 131–144. (doi:10.1152/jn.00872.2006)

Tsukamoto N, Nagaya T, Kuwayama A, Takano K, Shizume K, Sugita K & Seo H 1994 Octreotide treatment results in the inhibition of GH gene expression in the adenoma of the patients with acromegaly. *Endocrine Journal* **41** 437–444. (doi:10.1507/endocrj.41.437)

Tsukamoto N, Otsuka F, Miyoshi T, Yamanaka R, Inagaki K, Yamashita M, Otani H, Takeda M, Suzuki J, Ogura T *et al.* 2010 Effects of bone morphogenetic protein (BMP) on adrenocorticotropin production by pituitary corticotrope cells: involvement of up-regulation of BMP receptor signaling by somatostatin analogs. *Endocrinology* **151** 1129–1141. (doi:10.1210/en.2009-1102)

Tsukamoto N, Otsuka F, Miyoshi T, Inagaki K, Nakamura E, Suzuki J, Ogura T, Iwasaki Y & Makino H 2011 Activities of bone morphogenetic proteins in prolactin regulation by somatostatin analogs in rat pituitary GH3 cells. *Molecular and Cellular Endocrinology* **332** 163–169. (doi:10.1016/j.mce.2010.10.008)

Tyrrell JB, Lorenzi M, Gerich JE & Forsham PH 1975 Inhibition by somatostatin of ACTH secretion in Nelson's syndrome. *Journal of Clinical Endocrinology and Metabolism* **40** 1125–1127. (doi:10.1210/jcem-40-6-1125)

Vesely DL 1980 The interrelationship of somatostatin and guanylate cyclase activity. *Molecular and Cellular Biochemistry* **32** 131–134. (doi:10.1007/BF00227439)

Visser-Wisselaar HA, Van Uffelen CJ, Van Koetsveld PM, Lichtenauer-Kaligis EG, Waaijers AM, Uitterlinden P, Mooy DM, Lamberts SW & Hofland LJ 1997 17-β-Estradiol-dependent regulation of somatostatin receptor subtype expression in the 7315b prolactin secreting rat pituitary tumor *in vitro* and *in vivo*. *Endocrinology* **138** 1180–1189. (doi:10.1210/endo.138.3.5016)

Volpi R, Chiodera P, Capretti L, Caiazza A, Caffarri G, Magotti MG, Boni S & Coiro V 1996 Inhibition by somatostatin of the growth hormone, but not corticotropin response to angiotensin II in normal men. *Hormone Research* **45** 269–272. (doi:10.1159/000184804)

War SA & Kumar U 2012 Coexpression of human somatostatin receptor-2 (SSTR2) and SSTR3 modulates antiproliferative signaling and apoptosis. *Journal of Molecular Signaling* **7** 5. (doi:10.1186/1750-2187-7-5)

Werle M & Bernkop-Schnurch A 2006 Strategies to improve plasma half life time of peptide and protein drugs. *Amino Acids* **30** 351–367. (doi:10.1007/s00726-005-0289-3)

Werner H, Koch Y, Baldino F Jr & Gozes I 1988 Steroid regulation of somatostatin mRNA in the rat hypothalamus. *Journal of Biological Chemistry* **263** 7666–7671.

White RE, Schonbrunn A & Armstrong DL 1991 Somatostatin stimulates Ca(2+)-activated K^+ channels through protein dephosphorylation. *Nature* **351** 570–573. (doi:10.1038/351570a0)

White RE, Lee AB, Shcherbatko AD, Lincoln TM, Schonbrunn A & Armstrong DL 1993 Potassium channel stimulation by natriuretic peptides through cGMP-dependent dephosphorylation. *Nature* **361** 263–266. (doi:10.1038/361263a0)

Xu Y, Berelowitz M & Bruno JF 1995 Dexamethasone regulates somatostatin receptor subtype messenger ribonucleic acid expression in rat pituitary GH4C1 cells. *Endocrinology* **136** 5070–5075. (doi:10.1210/endo.136.11.7588243)

http://jme.endocrinology-journals.org
DOI: 10.1530/JME-14-0034

Published by Bioscientifica Ltd.

Yan M, Jones ME, Hernandez M, Liu D, Simpson ER & Chen C 2004 Functional modification of pituitary somatotropes in the aromatase knockout mouse and the effect of estrogen replacement. *Endocrinology* **145** 604–612. (doi:10.1210/en.2003-0646)

Yang SK & Chen C 2007 Involvement of somatostatin receptor subtypes in membrane ion channel modification by somatostatin in pituitary somatotropes. *Clinical and Experimental Pharmacology & Physiology* **34** 1221–1227. (doi:10.1111/j.1440-1681.2007.04806.x)

Yang SK, Parkington HC, Blake AD, Keating DJ & Chen C 2005 Somatostatin increases voltage-gated K^+ currents in GH3 cells through activation of multiple somatostatin receptors. *Endocrinology* **146** 4975–4984. (doi:10.1210/en.2005-0696)

Yang SK, Parkington HC, Epelbaum J, Keating DJ & Chen C 2007 Somatostatin decreases voltage-gated Ca^{2+} currents in GH3 cells through activation of somatostatin receptor 2. *American Journal of Physiology. Endocrinology and Metabolism* **292** E1863–E1870. (doi:10.1152/ajpendo.00047.2007)

Yu WH, Kimura M & McCann SM 1997 Effect of somatostatin on the release of gonadotropins in male rats. *Proceedings of the Society for Experimental Biology and Medicine* **214** 83–86. (doi:10.3181/00379727-214-44073)

Zatelli MC, Piccin D, Tagliati F, Ambrosio MR, Margutti A, Padovani R, Scanarini M, Culler MD & degli Uberti EC 2003 Somatostatin receptor subtype 1 selective activation in human growth hormone (GH)- and prolactin (PRL)-secreting pituitary adenomas: effects on cell viability, GH, and PRL secretion. *Journal of Clinical Endocrinology and Metabolism* **88** 2797–2802. (doi:10.1210/jc.2002-021825)

Zatelli MC, Piccin D, Vignali C, Tagliati F, Ambrosio MR, Bondanelli M, Cimino V, Bianchi A, Schmid HA, Scanarini M *et al*. 2007 Pasireotide, a multiple somatostatin receptor subtypes ligand, reduces cell viability in non-functioning pituitary adenomas by inhibiting vascular endothelial growth factor secretion. *Endocrine-Related Cancer* **14** 91–102. (doi:10.1677/ERC-06-0026)

Zeyda T & Hochgeschwender U 2008 Null mutant mouse models of somatostatin and cortistatin, and their receptors. *Molecular and Cellular Endocrinology* **286** 18–25. (doi:10.1016/j.mce.2007.11.029)

Zeyda T, Diehl N, Paylor R, Brennan MB & Hochgeschwender U 2001 Impairment in motor learning of somatostatin null mutant mice. *Brain Research* **906** 107–114. (doi:10.1016/S0006-8993(01)02563-X)

Zhang L, Li W, Hong X & Lin H 2009 Regulation of preprosomatostatin 1 (PSS1) gene expression by 17beta-estradiol and identification of the PSS1 promoter region in orange-spotted grouper (Epinephelus coioides). *Molecular and Cellular Endocrinology* **311** 87–93. (doi:10.1016/j.mce.2009.06.008)

Zheng H, Bailey A, Jiang MH, Honda K, Chen HY, Trumbauer ME, Van der Ploeg LH, Schaeffer JM, Leng G & Smith RG 1997 Somatostatin receptor subtype 2 knockout mice are refractory to growth hormone-negative feedback on arcuate neurons. *Molecular Endocrinology* **11** 1709–1717. (doi:10.1210/mend.11.11.0016)

Zorrilla R, Simard J, Rheaume E, Labrie F & Pelletier G 1990 Multihormonal control of pre-pro-somatostatin mRNA levels in the periventricular nucleus of the male and female rat hypothalamus. *Neuroendocrinology* **52** 527–536. (doi:10.1159/000125639)

Received in final form 17 April 2014
Accepted 24 April 2014
Accepted Preprint published online 29 April 2014

http://jme.endocrinology-journals.org
DOI: 10.1530/JME-14-0034

Published by Bioscientifica Ltd.