Does pasireotide directly modulate skeletal muscle metabolism?

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Cushing's Disease (CD) is a severe endocrine disease caused by corticotroph pituitary adenomas. CD is associated to high mortality primarily dependent on cardiovascular comorbidities induced by glucocorticoid (GC) excess-related metabolic changes [1].

Besides a number of other metabolic comorbidities, such as hyperglycemia, dyslipidaemia, and obesity, protein metabolism is severely affected by GC excess via direct and indirect stimulation of protein degradation and inhibition of protein synthesis, which can lead to muscle tissue loss [2].

Pasireotide, a stable cyclohexapeptide showing high affinity for multiple somatostatin receptor (SSTR) subtypes (sst5 > sst2 > sst3 > sst1), currently represents the only pituitary-targeted drug approved for clinical use in CD by both European Medicines Agency and Food and Drug administration [1, 3]. However, despite the initial search for a compound able to closely mimicking native somatostatin (SRIF-14), preclinical studies have demonstrated that this compound shows different functional properties compared to both native somatostatin-14 and first-generation somatostatin analogs (SSA) (e.g., octreotide) [4]. In this context, due to the peculiar pharmacological characteristics of pasireotide, a number of studies investigated its potential role on extra-pituitary cell types and tissues [5, 6].

In light of the well-established presence of myopathy in Cushing's patients (muscle atrophy and weakness),

Tulipano and co-workers evaluated whether pasireotide might exert direct effects on skeletal muscle cells. For this purpose, the Authors investigated the effects of pasireotide in the modulation of protein turnover and its interaction with the synthetic glucocorticoid dexamethasone (DEX) in differentiated rat myoblast-like cells (L6 cell line), a model of skeletal muscle cells [7]. More in detail, they focused on the effects of pasireotide on the mTOR-p70S6 kinase-S6 ribosomal protein signaling pathway, which plays a crucial role in the protein synthesis machinery [2]. From a clinical perspective, the demonstration of a direct anabolic effect of pasireotide on muscle cells could further strengthen the rationale for its use in CD treatment, and pave the way for pasireotide treatment in other diseases characterized by an excess of catabolism and loss of lean mass.

Interestingly, in differentiated L6 cells, Tulipano and co-workers observed that pasireotide was able to increase protein synthesis (evaluated as [³H]-tyrosine incorporation), at the highest concentration tested (1 μM). However, pre-treatment with high dose of DEX (100 nM, 48 h) did not cause a significant decline of protein synthesis and did not affect the cell response to pasireotide [7]. On the other hand, DEX treatment significantly increased protein degradation, but pasireotide, that per se did not affect protein degradation, did not reverse the activity of DEX in this specific cell model, as well [7].

As we mentioned above, loss of muscle and lean mass depends on both protein synthesis and protein degradation. In their experimental setting, the Authors observed a positive effect of pasireotide on protein synthesis, and therefore they further investigated one of the main intracellular pathways related to this aspect. Indeed, during GC-mediated reduction of protein synthesis, GCs interfere with the

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stimulatory activity of hormones (e.g., insulin, IGF-1) and amino acids on Akt-mTOR pathway and, subsequently, on the phosphorylation of their substrates, such as the S6 ribosomal protein [2].

In this context, the Authors observed that pasireotide time dependently enhanced the stimulatory effect of serum on p70S6K phosphorylation, but did not change the levels of S6 ribosomal protein phosphorylation (the p70S6K substrate). The Authors interpreted this evidence suggesting that pasireotide is not able to directly induce the activation of the p70S6K-S6 ribosomal protein pathway in differentiated L6 cells.

This finding, although in contrast with the effect of high-dose pasireotide in increasing protein synthesis reported in this study, is in line with the previously demonstrated role of SSAs in the inhibition of mTOR and its effectors p70/S6K and 4E-BP1 in different cell models [6, 8]. Therefore, as suggested by the Authors, the Akt-mTOR pathway does not seem to be involved in the pasireotide-mediated anabolic effects, and other signaling cascades could mediate this drug-related activity. Noteworthy, a number of pre-clinical studies already demonstrated that pasireotide is able to activate a number of different effectors in normal and tumor cells, modulating cell functions in both a direct and indirect way. For example, pasireotide has been shown to exert its anti-proliferative effect via either the down-regulation of MAP kinase pathway, or the inhibition of growth or angiogenic factors [6, 8, 9].

Furthermore, a puzzling observation derived by the study conducted by Tulipano and co-workers is that the observed activity is not mediated by canonical SSTRs, since differentiated L6 cells do not express SSTRs. However, although it is widely (and reasonably) assumed that SSA activity results from the correspondence between its binding profile and the pattern of SSTR expression in the target cells, other evidences suggest that additional mechanisms, distinct from the mere presence of a given receptor, are needed to achieve the functional effect of the compound [10]. In particular, this seems to be the situation of pasireotide, whose functional and pharmacological properties are far to be fully understood. In any case, the Authors concluded that their “study do not support a direct pasireotide effect on skeletal muscle”.

In this respect, we want to remark the importance, among the scientific community, of the publication of the so-called “negative data”. Although this attitude has been openly encouraged in the recent years, and nowadays several clinical trials present data demonstrating the failure of the primary hypothesis, still few scientific papers report results not confirming the authors’ starting assumptions. This is particularly evident in basic and translational research. However, as demonstrated by this report, negative data can provide the scientific community with important

information and drive future research, when supported by a strong starting hypothesis. Possibly, L6 cells are not the best model to investigate the effect of pasireotide on protein turnover, and being pasireotide-mediated effect on protein synthesis not mediated by Akt-mTOR pathway, other mechanisms need to be explored. All this information derives from “conflicting results”, as stated by the Authors themselves.

However, they can represent a precious starting point for future studies investigating the role of pasireotide on muscle cells, protein turnover and, more in general, extra-pituitary tissues.

Compliance with Ethical Standards

Conflict of Interest F.G. received fees for lectures and/or participation to advisory boards for Novartis, AMCo, and IONIS Pharmaceuticals, T.F. declares that he has no conflict of interest.

Ethical approval This article does not contain any studies with human participants performed by any of the authors.

References

1. R. Pivonello, M. De Leo, A. Cozzolino, A. Colao, The treatment of Cushing's disease. *Endocr. Rev.* **36**(4), 385–486 (2015). doi:[10.1210/er.2013-1048](https://doi.org/10.1210/er.2013-1048)
2. F. Ferrau, M. Korbonits, Metabolic comorbidities in Cushing's syndrome. *Eur. J. Endocrinol.* **173**(4), M133–M157 (2015). doi:[10.1530/EJE-15-0354](https://doi.org/10.1530/EJE-15-0354)
3. C. Bruns, I. Lewis, U. Briner, G. Meno-Tetang, G. Weckbecker, SOM230: a novel somatostatin peptidomimetic with broad somatotropin release inhibiting factor (SRIF) receptor binding and a unique antiseecretory profile. *Eur. J. Endocrinol.* **146**(5), 707–716 (2002)
4. R. Cescato, K.A. Loesch, B. Waser, H.R. Macke, J.E. Rivier, J.C. Reubi, A. Schonbrunn, Agonist-biased signaling at the sst2A receptor: the multi-somatostatin analogs KE108 and SOM230 activate and antagonize distinct signaling pathways. *Mol. Endocrinol.* **24**(1), 240–249 (2010). doi:[10.1210/me.2009-0321](https://doi.org/10.1210/me.2009-0321)
5. D.L. Kleinberg, P. Ameri, B. Singh, Pasireotide, an IGF-I action inhibitor, prevents growth hormone and estradiol-induced mammary hyperplasia. *Pituitary* **14**(1), 44–52 (2011). doi:[10.1007/s11102-010-0257-0](https://doi.org/10.1007/s11102-010-0257-0)
6. S. Moatassim-Billah, C. Duluc, R. Samain, C. Jean, A. Perraud, E. Decaup, S. Cassant-Sourdy, Y. Bakri, J. Selves, H. Schmid, Y. Martineau, M. Mathonnet, S. Pyronnet, C. Bousquet, Anti-metastatic potential of somatostatin analog SOM230: Indirect pharmacological targeting of pancreatic cancer-associated fibroblasts. *Oncotarget* **7**(27), 41584–41598 (2016). doi:[10.18632/oncotarget.9296](https://doi.org/10.18632/oncotarget.9296)
7. G. Tulipano, L. Faggi, S. Schulz, M. Spinello, A. Giustina, Effects of pasireotide (SOM230) on protein turnover and p70S6 kinase-S6 ribosomal protein signaling pathway in rat skeletal muscle cells. *Endocrine* (2017). doi:[10.1007/s12020-017-1235-3](https://doi.org/10.1007/s12020-017-1235-3)
8. M. Theodoropoulou, G.K. Stalla, Somatostatin receptors: from signaling to clinical practice. *Front. Neuroendocrinol.* **34**(3), 228–252 (2013). doi:[10.1016/j.yfrne.2013.07.005](https://doi.org/10.1016/j.yfrne.2013.07.005)
9. M.C. Zatelli, D. Piccin, C. Vignali, F. Tagliati, M.R. Ambrosio, M. Bondanelli, V. Cimino, A. Bianchi, H.A. Schmid, M. Scanarini, A. Pontecorvi, L. De Marinis, G. Maira, E.C. degli Uberti,

- Pasireotide, a multiple somatostatin receptor subtypes ligand, reduces cell viability in non-functioning pituitary adenomas by inhibiting vascular endothelial growth factor secretion. *Endocr. Relat. Cancer* **14**(1), 91–102 (2007). doi:[10.1677/ERC-06-0026](https://doi.org/10.1677/ERC-06-0026)
10. A. Ibanez-Costa, E. Rivero-Cortes, M.C. Vazquez-Borrego, M.D. Gahete, L. Jimenez-Reina, E. Venegas, A. de la Riva, M.A. Arraez, I. Gonzalez-Molero, H.A. Schmid, S. Maraver-Selfa, I. Gavilan-Villarejo, J.A. Garcia-Arnes, M.A. Japon, A. Soto, M.A. Galvez, R.M. Luque Huertas, J.P. Castano, Octreotide and Pasireotide (dis)similarly inhibit pituitary tumor cells in vitro. *J. Endocrinol.* (2016). doi:[10.1530/JOE-16-0332](https://doi.org/10.1530/JOE-16-0332)