

Title: Intracellular behavior of the novel sympathetic nerve agent ^{18}F -LMI1195

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Introduction: ^{18}F -N-[3-bromo-4-(3-fluoro-propoxy)-benzyl]-guanidine (^{18}F -LMI1195) is a novel PET tracer for the assessment of sympathetic cardiac nerve conditions. Although the desirable image quality with high neural uptake has been demonstrated, the intracellular behavior is not yet fully understood. We aimed to elucidate whether ^{18}F -LMI1195 is stored in storage vesicles and released in accompany with vesicle turnover.

Methods: Two different kinds of norepinephrine transporter-expressing cell lines were used for in vitro tracer uptake studies: PC12 (rat pheochromocytoma) and SK-N-SH (human neuroblastoma). After 2 h of ^{18}F -LMI1195 loading into PC12 cells, the medium was removed and the cells were washed twice. Then tracer release in response to reserpine (50 nM) or high potassium (100 mM) treatment, as a stimulant for storage vesicle turnover, were evoked at 37 °C for 10, 20, and 30 min. After these treatments, 500 μL of buffer was collected to determine radioactivity present in the extracellular medium. As a reference, ^{131}I -MIBG uptake was also examined in the same manner.

Results: In PC12 cells, reserpine induced an increase in ^{18}F -LMI1195 release compared to controls (163 \pm 8 % of control release at 30 min). Treatment with high potassium also promoted the release (515 \pm 23 % of control release at 30 min) while this release induction effect was inhibited by chelation of extracellular Ca^{2+} with EDTA (164 \pm 29 % of control release at 30 min). These findings indicate that tracer release resulted from Ca^{2+} influx, which caused by membrane depolarization by high potassium. In contrast, both treatments had no effect on ^{18}F -LMI1195 release in SK-N-SH cells, that lack catecholamine storage vesicles. Similar tracer kinetics after reserpine treatments were confirmed with ^{131}I -MIBG.

Conclusion: In vitro tracer uptake study confirmed both ^{18}F -LMI1195 and ^{131}I -MIBG are stored in vesicles after transported into cells, and released along with storage vesicle turnover. Understanding the underlying kinetics of ^{18}F -LMI1195 at the subcellular level would help

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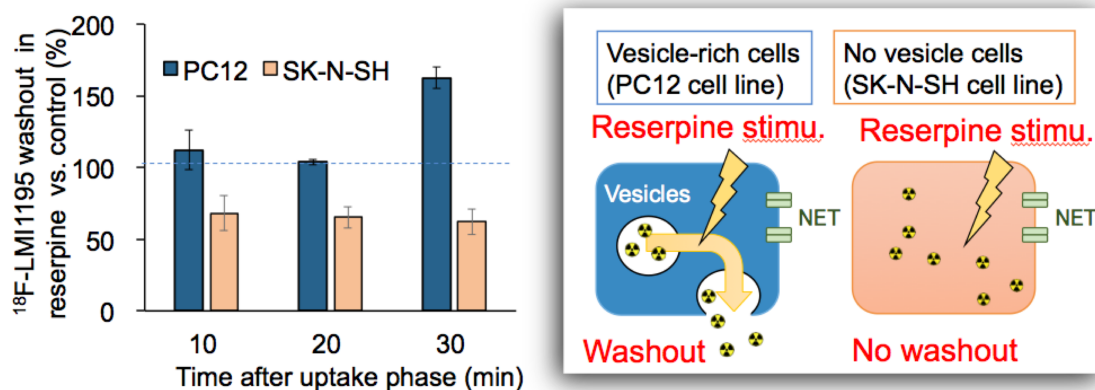


Figure 1. Effect of reserpine treatment on ¹⁸F-LMI1195 washout from PC12 and SK-N-SH cells. Reserpine-induced depletion of stored tracers can be occurred only in cells with storage vesicles.

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