THE SYNTHESIS AND BIODISTRIBUTION OF A RADIOIODINATED GAMMAGLOBULIN DERIVATIVE

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INTRODUCTION

The development of two tracers, N-isopropyl-p-(I123)-iodoamphetamine (I123-IMP) (1,2) and N, N, N',N'-trimethyl-N'-(2-hydroxyl-3-methyl-5-iodobenzyl)-1,3-propanediamine(I123)(I123-HIPDM) (3) has resulted in a renewed interest within the realm of nuclear medicine in the measurement of cerebral blood flow. Of particular interest is the use of these radiopharmaceuticals for the early diagnosis of stroke. Using emission tomography with I123-IMP, Hill and co-workers (4) were able to detect regions of cerebral impairment prior to their appearance as abnormalities on CT scans.

It is important to bear in mind that I123-IMP and I123-HIPDM distribute in the brain in proportion to cerebral blood flow and thus delineate regions of vascular insult indirectly; that is, infarcts appear as photon-deficient areas within a "hot" background. Contamination of the image by scattered photons, especially those resulting from the high-energy emissions of I124 impurities, and the superimposition of activity from normal brain tissue in adjacent planes, both make it difficult to define the boundaries of a lesion unless tomographic instrumentation is employed.

The development of a radiopharmaceutical which accumulates preferentially in damaged brain tissue might provide a more sensitive and selective method for the early detection of cerebral infarcts. Ganglioside G\textsubscript{M1} is a glycosphingolipid which appears to be involved in the repair process of damaged nerve tissue (5-7). When labelled with an isotope such as I123, G\textsubscript{M1} might be useful as an infarct-avid tracer for the early diagnosis of stroke. The purpose of this study was...
first to develop a rapid and efficient technique for labelling $G_{M1}$ with $I^{125}$. The stability and neuronal membrane binding properties were then studied in vitro. Finally the distribution of $I^{125}$ following injection of $I^{125}$ labelled $G_{M1}$ was studied in the mouse and, in preliminary fashion, in the gerbil infarct model. A more detailed description of the in vivo aspects of this work will appear elsewhere (8).

METHODS OF PROCEDURE

Labelling $G_{M1}$ with $I^{125}$. Direct iodination. Ganglioside $G_{M1}$ isolated from bovine brain by the method of Tettamanti and co-workers (9) was obtained from FIDIA Laboratories. Initial attempts to label $G_{M1}$ with $I^{125}$ were performed using the iodine monochloride method (10), modified as follows: The $I^{125}$ activity (1 mCi, adjusted to pH 7.4) was added to 0.5 ml methanol/chloroform (1:1) containing 16 $\mu$mol of $G_{M1}$. Addition of 1.6 $\mu$mol each of $I_2$ and HgCl$_2$, both in CH$_3$OH/CHCl$_3$, generated the ICI in situ. The reaction was terminated after 12 hr by the addition of an excess of sodium metabisulfite. After drying under nitrogen, the reaction mixture was resuspended in phosphate buffered saline (pH 7.4) and passed through a Sephadex G-50 column to separate the I$^{125}$ labelled $G_{M1}$ from inorganic I$^{125}$.

Radioiodination of a $G_{M1}$-tyramine conjugate. A method analogous to that reported by Klemm and co-workers (11) was used to synthesize the $G_{M1}$-tyramine conjugate. Oxidation of the terminal galactose residue of $G_{M1}$ was effected by addition of 30 $\mu$mol of KIO$_4$ to 20 $\mu$mol of $G_{M1}$ in 0.01 M pH 8.4 borate buffer. The reaction was terminated after 4 hr at 37°C by the addition of an excess of glycerol. The oxidized $G_{M1}$ micelles were purified by dialysis against pH 8.4 borate buffer and then reacted for 24 hr at 25°C with a 4-fold molar excess of both tyramine and 2 M NaBH$_3$CN in pH 8.4 borate. The reaction mixture was then dialyzed against borate buffer for 48 hr.

The $G_{M1}$-tyramine conjugate was labelled with $I^{125}$ using iodogen (12). Varying amounts of iodogen in methylene chloride were added to 5-dram glass vials and evaporated under nitrogen.