EDITORIAL

Myocardial Metabolism of Radioiodinated BMIPP

In this issue of *The Journal of Nuclear Medicine*, Yamamichi et al. (*1*) describe the isolation and identification of catabolites from the outflow of a recirculating isolated rat heart system following administration of radioiodinated 15-(p-iodophenyl)-3-R,S-methylpentadecanoic acid (BMIPP) and the effects of perfusate substrates on metabolite formation. BMIPP is a modified fatty acid analogue of [15-(p-iodophenyl-) pentadecanoic acid (IPPA) in which a methyl group has been introduced into the beta (3)-position of the fatty acid chain to inhibit rapid myocardial catabolism (*2,3*). Increasing interest in the clinical use of ^{123}I-labeled fatty acids for myocardial imaging is illustrated by the broad use of ^{123}I-BMIPP in Japan, following its commercial introduction as Cardiodine by Nihon Medi-Physics Co., Inc., in 1993. The identification of the metabolites released from the myocardium described by Yamamichi et al. is important in further expanding our understanding of the BMIPP metabolism and the factors which may affect catabolism and myocardial washout of this important cardiac imaging agent.

Clinical use of ^{123}I-labeled modified fatty acid analogs which show delayed myocardial clearance for myocardial SPECT imaging is based on differences between regional myocardial fatty acid uptake patterns and flow tracer distribution which are often observed in various myocardial disorders (*2,3*). The use of ^{123}I-BMIPP in conjunction with flow tracers may represent a unique opportunity to correlate energy substrate metabolism with regional myocardial viability using SPECT. Although the physiological basis is not completely understood, differences between regional fatty acid and flow tracer distribution may indirectly reflect alterations in important parameters of metabolism which can be important for clinical decision making.

RATIONALE FOR DEVELOPING IODINE-123-LABELED METHYL-BRANCHED FATTY ACIDS

Naturally occurring, long-chain fatty acids (palmitic, oleic, etc.) are the principal energy source for the normoxic myocardium, and thus, radioiodinated fatty acid agents represent potential probes to evaluate differences in cardiac oxidative metabolism which are present in various myocardial disorders. For cardiac SPECT, especially with single- and double-headed cameras, significant tracer redistribution during the acquisition period must be minimized during the relatively long time periods required for camera rotation and data collection. Since straight chain fatty acids are rapidly metabolized by beta-oxidation, a variety of structural modifications have been introduced into fatty acid analogs to evaluate their inhibition of beta-oxidation, thus delaying myocardial tracer clearance and relative regional redistribution. In particular, methyl-branched fatty acids such as BMIPP have been widely investigated (*2–6*). The effects of methyl-substitution in delaying myocardial tracer clearance is based on the expected inhibition of the beta-oxidative process at the stage of obligatory NAD-dependent oxidation of the beta-hydroxy intermediate catalyzed by beta-hydroxyacyl-CoA dehydrogenase.

During the normal oxidative catabolism of aliphatic (straight chain) fatty acids, oxidation forms the beta-ketoacyl-CoA intermediate, which is the substrate for thiolase-catalyzed alpha, betacarbon-carbon bond cleavage with the release of the acetyl CoA two-carbon fragment. The chain length of the original fatty acid substrate is thus decreased by two carbon atoms and the resulting acyl-CoA product can then be recycled through the beta-oxidative chain.

The potential clinical use of radioiodinated fatty acids for cardiac imaging was first demonstrated in 1965 with ^{131}I-labeled iodo-oleic acid (*7*). For nearly 30 yr, use of radiolabeled fatty acids for myocardial imaging has fascinated many investigators (*2–6*). Development of the BMIPP analog at the Oak Ridge National Laboratory (ORNL) with radioiodine stabilized by attachment to the para-position of the terminal phenyl ring (*8–11*) was based on introduction of 3-methyl-branching into IPPA. The inhibitory effect of the 3-methyl group of BMIPP on beta-oxidation results in longer myocardial retention, which has been demonstrated in extensive in vivo studies in laboratory animal species and, more recently, in humans (*2,3*).

Oxidative Products Formed from BMIPP

The results of Yamamichi et al. have for the first time identified the metabolites released from the myocardium following administration of BMIPP. As proposed earlier (*12–15*), the 3-methyl group is expected to interfere with betadation, but this impediment could be removed by initial alpha-oxidation, and the resulting alpha-methyl-substituted metabolite could then serve as a substrate for the usual beta-oxidative pathway. Consistent with this expectation, the principal metabolite identified by Yamamichi et al. is the initial alpha-oxidation product 14-(p-iodophenyl)-2(α)-R,S-methyltetradecanoic acid (AMIPT).

Other metabolic products which were also identified by comparison with synthetic standards are 12-(p-iodophenyl)dodecanoic acid ($PIPC_{12}$) and 2-(p-iodophenyl)acetic acid (PIPA). BMIPP is thus apparently metabo-

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lized, as had been expected, by initial alpha-oxidation in which the carboxyl carbon is lost. Since the impediment of the beta-methyl substituent has been removed, the alpha-methyl-substituted CoA fragment can then pass through the beta-oxidative chain, with p-iodophenylacetic acid being formed as the final oxidative product, which is a process similar to the oxidative metabolism of phytanic acid which is inhibited in Refsum's disease (*9*). An important aspect of this scenario which is yet unanswered, however, is the intracellular dynamics of the activated BMIPP species, since it has not yet been established if initial alpha-oxidation occurs in the mitochondrial compartment. Initial alpha-oxidation of BMIPP CoA in an extramitochondrial compartment would be expected to require subsequent transfer via the carnitine shuttle through the mitochondrial membrane for beta-oxidative catabolism.

Earlier studies had used both nonworking (*13,14*) and working (*15*) rat hearts to isolate an unidentified polar metabolite from the outflow of Langendorff preparations. In these studies, only one radioactive component was observed by thin-layer (TLC) and high-performance liquid chromatographic (HPLC) studies, and the same single metabolite was also observed in plasma samples from humans following intravenous administration of [^{123}I]BMIPP (*2,3,16,17*). Although the identification of only one polar metabolite in earlier isolated heart studies, but several catabolites by Yamamichi et al. may result from different experimental conditions, this does not explain the single component observed in earlier patient plasma analysis (*16,17*). It is important to further extend the metabolite studies now that authentic samples are available.

The studies by Yamamichi et al. are important since they provide one missing link in our understanding of the intracellular metabolism of BMIPP and the identity of the catabolic species which immediately leaves the cell. Although the intracellular accumulation of BMIPP metabolites was not evaluated by Yamamichi et al. an important observation of earlier work (*13–18*) was that the oxidative catabolic products from BMIPP do not accumulate intracellularly and evidently immediately leave the cell after formation. Thus, the oxidative catabolites from BMIPP do not accumulate in myocytes and if present in myocardial lipid extracts must be present in very small quantities.

Intracellular Metabolism of BMIPP

Although the oxidative catabolite(s) of BMIPP had not been identified until the work by Yamamichi et al., the identity of the complex lipid products accumulating in the myocytes from metabolism of BMIPP are well understood and have been studied in detail (*2,3*). High-performance liquid chromatography has been used to examine the incorporation of radioiodinated BMIPP and IPPA into intracellular phospholipid fractions of myocardial lipids isolated from both rats and dogs. Structural modifications have unexpected effects on fatty acid metabolism. The IPPA straight-chain analog is incorporated into lecithin (phosphatidylcholine) in rat hearts, while BMIPP is primarily found in the cephalin (phosphatidylethanolamine) fraction (*17*). Physiological factors which affect myocardial uptake of BMIPP have been widely studied (*2,3*) and include the effects of ATP (*19,20*).

The correlation between myocardial ATP levels and BMIPP uptake could be especially important and this relationship has been investigated in normal and salt-sensitive Dahl-strain rats following administration of an inhibitor of mitochondrial carnitine acyltransferase I (tetradecylglycidic acid (TDGA) (*20*). Initial myocardial BMIPP uptake was not influenced by acute inhibition of beta-oxidation by the inhibitor. Intracellular ATP levels correlating with BMIPP retention are interpreted to reflect cystolic activation of BMIPP to BMIPP-CoA with slow shunting into triglyceride storage products. In the hypertrophied hearts of Dahl rats, a negative correlation was observed between ATP levels and severely compromised myocardial BMIPP. Differences in mitochondrial and cytosolic ATP pools may explain these results, but additional studies are required to more fully evaluate the relationship between high-energy phosphate levels and BMIPP retention. Studies have pursued evaluation of the influence of methyl-substitution on the myocardial metabolism of BMIPP in rat hearts in vivo (*21*) and isolated Langendorff-perfused rat hearts (*13–15*), which demonstrated incorporation of BMIPP into triglyceride storage products correlating with the observed myocardial retention.

Mismatch of Iodine-123-BMIPP and Flow Tracer Myocardial Distribution

The apparently unique, yet not well understood, property of BMIPP is the mismatch often observed between the regional distribution of this agent and flow tracers, although it has not yet been established if these differences are even related to the intracellular oxidative metabolism of BMIPP. Initial studies by Yonekura et al. and Yamamoto et al. clearly demonstrated differences in BMIPP and flow tracer distribution in the hearts of hypertensive animals using high-resolution autoradiographic techniques (*22,23*), and these important studies have relevance to the clinical applications of [^{123}I]BMIPP.

Animal studies with radioiodinated BMIPP have also evaluated the regional myocardial distribution of this tracer in various cardiac disease models and paved the way for subsequent human studies. Significant differences have been found between flow tracer (^{201}Tl) and methyl-branched fatty acid distribution in the free wall of the left ventricle and septal regions of hearts from hypertensive rats (*22,23*) and rat and hamster models with hypertrophic and cardiomyopathic heart disease (*24,25*). BMIPP also has been used to evaluate cocaine-induced regional myocardial metabolic changes in hypertensive rats by comparison of regional perfusion of ^{201}Tl with differences in 2-deoxyglucose (2-DG) and radioiodinated BMIPP uptake (*26,27*).

Early studies have also evaluated the properties of radioiodinated BMIPP in an ischemic canine model (*28*) and have shown differences in BMIPP versus ^{201}Tl uptake in border zone regions.

The uptake and clearance of ^{123}I-BMIPP has been recently studied by planar imaging in a canine occlusion-reperfusion model (*29*) using ex vivo gamma camera imaging of the excised hearts.

The results from these extensive animal studies with radioiodinated BMIPP have provided an important foundation for humans studies now in progress. Only the global release of catabolites from rats in vivo or isolated heart preparations has been studied and the slow myocardial washout observed after administration of radioiodinated BMIPP in vivo or with isolated heart preparations represents only a small proportion of the total uptake. Regional mismatches in BMIPP uptake in comparison with flow tracer distribution may thus be expected to result from other factors unrelated to oxidative catabolism. Furthermore, the factors resulting in the mismatch phenomena between BMIPP and flow tracers are probably unrelated to the formation of oxidative products. They are probably related to differences in cellular uptake in comparison to normal myocytes and those cells which are viable but in which fatty acid metabolism is somehow impaired.

SUMMARY

After significant efforts over the last 30 yr, the use of ^{123}I-labeled fatty acids for routine assessment of cardiac disease may now be a reality with ^{123}I-BMIPP. The use of ^{123}I-labeled fatty acids may provide complementary information on myocardial viability, for example, identifying and assessing the presence of salvageable tissue. The results of these extensive clinical trials often report the mismatch between BMIPP and flow tracer distribution. But why? It will be important to continue the basic studies such as those reported by Yamamichi et al. to obtain a complete understanding of the metabolism and physiological factors affecting myocardial uptake of BMIPP.

Factors requiring additional study in animal models include biochemical and histological analysis of biopsy segments from myocardial regions which have decreased fatty acid relative to flow tracer uptake. An important issue for evaluation is the relative metabolism of the isomers of BMIPP, since BMIPP is a racemic mixture of the 3R-methyl and 3S-methyl isomers. It is certainly possible that only one isomer is catabolized to the metabolites identified by Yamamichi et al. and that the second isomer could be metabolically inactive. In addition to an evaluation of the relative intracellular metabolism of both isomers, the kinetics of tissue uptake and release of metabolites of the 3R- and 3S-methyl isomers of radioiodinated BMIPP should be evaluated. It is conceivable for instance, that one isomer may have less nontarget tissue uptake, in comparison with racemic 3-(R,S)-BMIPP.

Clinical protocols are being pursued at several institutions in western Europe (*17,30–32*), and ^{123}I-labeled BMIPP (Cardiodine) is currently widely used in Japan as an approved radiopharmaceutical for the evaluation of impairments of myocardial fatty acid metabolism and for myocardial viability (*6,33–35*). More than 2500 patient studies per month are currently conducted with Cardiodine in Japan in over 300 hospital-based nuclear medicine departments. This is estimated to represent about 14% of all nuclear cardiology studies in Japan. Through December 1994 more than 50,000 patient studies had been completed.

The results of these studies, in conjunction with clinical studies being pursued at several institutions in Europe, will hopefully stimulate the use of Cardiodine in the United States. We expect that the extensive data being documented on the use of ^{123}I-BMIPP in various myocardial disorders and the comparison of these results with commonly used flow tracers will answer these questions and provide the basis by which the broader possible role of BMIPP in nuclear cardiology will be further assessed. Lipid analyses, after either coronary sinus sampling or studies of serial blood samples, obtained from patients following intravenous administration of ^{123}I-BMIPP may confirm that these same metabolites identified in the rat heart studies reported by Yamamichi et al. are formed in humans.

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