Review Article

PET Tracers and Radiochemistry

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Abstract

This paper provides a brief review of the radiochemistry of radiopharmaceuticals used in positron emission tomography (PET). It includes some history of PET, the basic formation of radionuclides in a cyclotron target, the processing of the precursor molecules into a useful PET radiotracer and the clinical significance and outlook for PET radiotracers. This review is based on a series of recent books and articles outlining the fundamental goals of PET and how radiochemistry plays a part in achieving these goals. It is also drawn from the literature that has been developed in PET over the last 30 years since PET became a research and valuable clinical tool. PET is a growing field and the clinical applications of the chemistry and technology have just begun to be explored. There is a great deal left to do in order to explore the full potential of PET in the clinic.

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Introduction

Positron emission tomography (PET) has become a powerful scientific and clinical tool for probing biochemical processes in the human body. This is due, in large part, to advances in instrumentation and synthetic chemistry. The clinical application of PET has mushroomed in the last decade and it has proven to be vital in the evaluation and diagnosis of disease. Measurement of altered biochemical pathways and metabolic levels *in vivo* in a non-invasive and often quantitative manner is done routinely. The broad scope, versatility and sensitivity of PET make it the most powerful molecular imaging technique currently available for clinical use.

There are 4 positron-emitting radioisotopes that are used more than any others. These are fluorine-18 (F-18), carbon-11 (C-11), nitrogen-13 (N-13) and oxygen-15 (O-15). The reason these are so commonly used is that they have many attractive properties, one of which is that they can be easily substituted directly into biomolecules. Substitution of C-11 for C-12 does not significantly alter the reaction time or mechanisms of a molecule. A similar situation exists for N-13 and O-15. F-18 can often be substituted for a hydroxy group on a molecule or placed in a position where its presence does not significantly alter the biological behaviour of the molecule.

When these short-lived PET radioisotopes are used, time is the critical parameter. In essence, PET radiotracers must be synthesised and imaged within a time frame compatible with the half-life of the isotope. For C-11, this typically amounts to about 10 minutes for isotope production, 40 minutes for radiotracer synthesis and up to about 90 minutes for PET imaging. Thus, the entire study must be orchestrated and carried out within about 2.5 hours. The synthesis of almost any PET radiotracer cannot take more than two half-lives. Taking longer than this will result in an unacceptable loss of specific activity that is often critical for PET studies.

This review is not meant to be exhaustive, but rather to give some of the most common methods for synthesis for the common PET radiopharmaceuticals and radiotracers, as well as a few examples of their uses in clinical practice.

History

PET has taken more than 40 years to progress from the first attempts to obtain an image using positron emitters to a clinically useful tool. The reason for this slow progress is the necessity for the development of several elements that had to merge into the imaging modality of today. Several early investigators demonstrated the advantage of positron imaging using coincidence-counting techniques. The first, and perhaps most prominent, was the work of Brownell et al[1,2] at Massachusetts General Hospitals. Their early work reported in 1953[1] used a rectilinear scanning technique, but

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by 1960[2] they had developed a clinically usable positron camera. Their camera produced mainly planar images, but did give some tomographic information. By late 1959, Kuhl and Edwards[3] had successfully accomplished transaxial emission tomography and by the late 1960s, they had developed the Mark II scanner.[4]

In the early 1960s, the Brookhaven National Laboratory group produced a true transaxial positron tomograph utilising a ring system of detectors that is highly reminiscent of modern tomographs.[5] The system gave poor results because the reconstruction methods used were inadequate. The earlier work on reconstruction methods was unknown to these pioneers.[6] The advancement of PET proceeded slowly until after the development of the advanced reconstruction techniques that accompanied the development of X-ray computed tomography (CT).[7] The more modern version of positron emission CT then became possible, and was first implemented by Phelps et al in the mid-1970s.[8] It must be recognised that the driving force behind the use of positron emitters centred on the availability of the radionuclides C-11, N-13, O-15 and F-18.

C-11 and F-18, the most commonly used radionuclides for PET, were discovered more than 60 years ago. The discovery of carbon-11 preceded that of carbon-14 by several years, so that it became the first radioactive isotope of carbon to be used for chemical and biochemical tracer studies prior to and during World War II.[9] Because of the extraordinary experimental limitations imposed by its 20.4-minute half-life, C-11 was largely replaced by C-14 (a half-life of 5730 years) which became available after World War II. Interest in C-11 and the 3 other short-lived positron emitters (F-18, N-13 and O-15; Table 1) was rekindled 2 decades later when it was appreciated that their short half-lives and body-penetrating photons provided the potential to image biochemical transformations in the living human body.

The successful synthesis and application of F-18 fluorodeoxyglucose by Wolf et al[10,11] in the mid-1970s provided another impetus for the advancement of PET. Once the broad utility of this tracer had been demonstrated by scientists in basic research, the medical community became excited by the possibilities and began to clamour for more clinical applications. Finally, PET could not advance to the status of a widely used clinical tool without the entry of commercial enterprises into the field. The manufacturer of sophisticated equipment for routine use demands major involvement if a modality is to gain widespread acceptance. That this has occurred is a tribute to the pioneers who foresaw the potential and carried out the basic studies that lead to clinical applications.

Precursor Production

The synthesis of all radiotracers in PET begins with the small precursor molecules that originate from the cyclotron target. The number of chemical forms of precursors for PET using the 4 traditional tracers is limited. This is a result of having to make the precursors in the rather harsh environment of the cyclotron target, where reactions take place between electronically and thermally excited atoms and molecules. In this environment, there is enough energy available to overcome most activation barriers and the final chemical form is often determined by the thermodynamics of the constituents. As a result, what in the target at the end of irradiation are very stable, unreactive molecules.

For C-11, the molecule that is the most stable in an oxidising environment is carbon dioxide (CO_2). In a reducing environment, the most stable is usually methane (CH_4). Either of these 2 chemical forms are the building blocks of more complex molecules. In the case of N-13, there are 2 common chemical forms. These are nitrogen gas (N_2) in the gas phase and nitrate (NO_3^-) in aqueous solution. NO_3^- is a desired form for further chemistry since N_2 is very unreactive under usual conditions. For O-15, the usual gaseous form is oxygen gas (O_2), which can be manipulated into other more useful chemical forms. In aqueous solution, the typical precursor is water. This is one of the precursors that can be used directly. In the case of F-18, the usual product out of the target is usually either fluoride ion (F^-) or fluorine gas (F_2), depending on the environment in the target during irradiation.

All of these radioisotopes can be produced from a wide variety of nuclear reactions, where the bombarding particle can be a proton, deuteron, helium-3 or helium-4. However, for each isotope there are only 1 or 2 reactions, usually with protons, that are commonly used. This is due mainly to the number of proton accelerators that are available in facilities where PET is done.

Table 1. Physical Characteristics of PET Radioisotopes

Nuclide	Half-life (min)	Decay mode	Maximum energy	Most prob. energy	Maximum range	Maximum specific activity (theoretical)
C-11	20.4	100% β^+	0.96 MeV	0.326 MeV	4.1 mm	9220 Ci/:mole
N-13	9.98	100% β^+	1.19 MeV	0.432 MeV	5.4 mm	18900 Ci/:mole
O-15	2.03	100% β^+	1.7 MeV	0.650 MeV	8.0 mm	91730 Ci/:mole
F-18	109.8	97% β^+ 3%EC	0.69 MeV	0.202 MeV	2.4 mm	1710 Ci/:mole

Carbon-11-Labelled Precursors

C-11 is most commonly produced in the target using the $^{14}N(p,\alpha)^{11}C$ nuclear reaction.[12] The target is usually a N_2 target with a trace of oxygen to convert C-11 into CO_2. Almost all syntheses involving C-11 start with CO_2 as the primary product. A number of other precursor molecules, some of which are shown in Figure 1, are synthesised from labelled CO_2, but all require some synthetic manipulation during or after cyclotron bombardment.

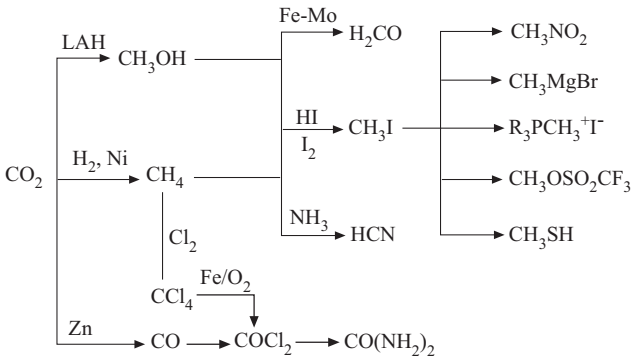

Fig. 1. The preparation of some common precursors for radiotracer synthesis from carbon dioxide.

Some of the earliest syntheses with carbon-11 depended directly on labelled CO_2 and labelled cyanide.[13] Today, however, alkylation with [^{11}C]methyl iodide is the most widely used method for introducing carbon-11 into organic molecules.[14]

CO_2 originating from the target can be easily and quickly processed into methane and methanol by simple reduction. The reduction is accomplished either by lithium aluminium hydride (which leads to methanol) or with hydrogen over a nickel catalyst (which leads to methane). Methyl iodide can be produced from both these precursors and there are commercial devices to carry out these syntheses. From methanol, hydrogen iodide is added, resulting in the production of methyl iodide. From methane, the gas is passed through a heated tube containing gaseous iodine and the methyl iodide is extracted in a recirculating system. Of special significance is an up-to-date listing, with references and structures, of most PET labelled compounds classified according to compound type.[15]

Specific activity is a critically important property in the preparation of radiotracers. Specific activity is the fraction of radiolabelled molecules relative to the total number of molecules and is usually expressed as a unit of radioactivity per mole of compound. It is particularly important in PET, where the radionuclide is incorporated into a radiotracer that is used to probe some physiological process, in which very small amounts of the native biomolecule are used. PET is basically a tracer method and the goal of the PET experiment is to probe the physiological process without perturbing that process. If the amount of radiotracer is very small, relative to the amount of the native compound or its competitor, then the process will be perturbed very little. When carrying out these studies, such as probing the number of receptors or the concentration of an enzyme, these considerations become even more important.[16]

There is, of course, an ultimate limit to specific activity when there are nothing but the radioactive atoms or radiolabelled molecules. The characteristics of the 4 PET isotopes are shown in Table 1.[17]

As an example, the typical specific activities for C-11-labelled molecules are in the order of 10 Curies/μmole (370 GBq/μmole). Hence, it can be seen that only1 in 1000 tracer molecules is actually labelled with C-11. The rest contain carbon-12.

Nitrogen-13 Labelled Precursors

N-13 is usually produced from 3 nuclear reactions which have become the standards. These are the $^{13}C(p,n)^{13}N$ reaction, usually carried out on isotopically enriched carbon powder,[18,19] the $^{12}C(d,n)^{13}N$ reaction carried out on natural carbon powder[20] and the $^{16}O(p,\alpha)^{13}N$ reaction, usually in water.[21]

As has been mentioned previously, N_2 is not a very useful synthetic precursor. The precursors most commonly used in the synthesis of N-13-containing compounds are nitrate ion from the water target and ammonia from the carbon powder targets. NO_3^- obtained from the water target can be easily converted into ammonia using a reduction with DeVarda's alloy[22] or with titanium chloride.[23]

Oxygen-15 Labelled Precursors

The usual reactions for the production of O-15 are the $^{15}N(p,n)^{15}O$ reaction on enriched N-15 or the $^{14}N(d,n)^{15}O$ reaction on natural nitrogen. The $^{16}O(p,pn)^{15}O$ reaction is also used when specific activity is not a concern. O-15 is usually in the form of O_2. If mixtures of nitrogen and oxygen are irradiated, the oxides of nitrogen can be produced directly;[24] if a mixture of hydrogen and oxygen are irradiated, labelled water will be produced.[25] These precursors can then be used to synthesise other oxygen-15 containing materials.

Fluorine-18 Labelled Precursors

F-18 is produced most often using 2 nuclear reactions.[26] The first is the $^{20}Ne(d,\alpha)^{18}F$ carried out in a neon gas target with added F_2 to keep the fluorine in an oxidised form. In this case, the fluorine is removed from the target as a gas mixture and can be used in the synthesis directly.[27] The second common reaction is the $^{18}O(p,n)^{18}F$ nuclear reaction on O-18-enriched water or oxygen-18-enriched O_2. There

are 2 separate cases for recovery of F-18 from these targets. For the gas target, the fluorine-18 can be recovered either as fluoride ion or as F_2. To recover F-18 as F_2, after irradiation O-18- enriched O_2 is removed and the target filled with a mixture of a trace amount of F_2 in an inert carrier gas such as argon.[28] The target is irradiated and the resulting mixture of $[^{18}F]F_2$ in argon is removed and used in synthesis directly. In the case of the water target, the activity is removed in the aqueous phase. There are 2 general methods after that. The first is to use the O-18 water containing [18F]fluoride ion directly in the synthesis. This method is used by several people who have small-volume water targets and the cost of losing the O-18 water is minor compared to the cost of the cyclotron run. The second method is to separate the fluoride from the O-18 water, either by distillation or by using a resin column.[29-31] When the resin is used, it also removes the metal ion impurities from the enriched fluoride solution which, in general, increases the reactivity of the fluoride. The fluoride can be made more reactive by combination with a metal ion complexing agent, such as a crown ether or a tetrabutylammonium salts.[32]

Common Synthetic Routes in PET

There are several excellent reviews of the chemistry involved in the preparation of radiopharmaceuticals.[15,17,33,34] Below are some specific examples and principles in synthetic chemistry using the short-lived positron emitters.

Carbon-11 Reactions

Alkylation with [11C]methyl iodide is the most widely used method for introducing carbon-11 into organic molecules since it was introduced in the late 1970s.[35,36] Alkylations are generally straightforward, as in the case of the synthesis of [11C]raclopride,[37] a widely used radiotracer for the dopamine D2 receptor that is synthesised by alkylating the nor-compound with [11C]methyl iodide.

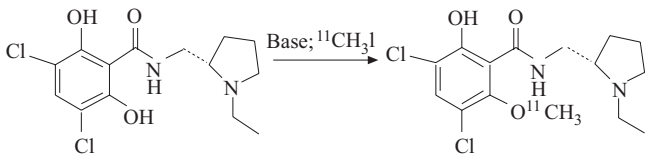

Fig. 2. Synthetic pathway for carbon-11 labelled raclopride.

Frequently, however, reactive centres on the substrate molecule must be shrouded with protective groups that can be rapidly removed.

C-11 synthesis is frequently complicated by the need for chiral-labelled products. In some cases, the chiral centre is present in the substrate and the reaction conditions preserve the chirality or, as an alternative, chiral high pressure liquid chromotography (HPLC) can be used to separate the desired labelled enantiomer from a labelled racemic mixture. However, asymmetric syntheses have also been developed and this technique has been used in the synthesis of labelled amines, ketones, aldehydes, acids and amino acids.[38]

Problems in C-11 synthesis generally include rigorously excluding stable carbon in order to maximise the specific activity of the product, optimising reaction rates and developing chromatographic methods that separate the labelled product from starting materials and by-products. Reaction times have been reduced and yields have been increased for many labelled compounds by applying microwave technology.[39]

C-11-labelled $^{11}CN^-$ has also been used extensively to obtain products containing carboxylic acids, primary amines or amide functionalities. This is done through the intermediate step of using a nitrile. The cyanide ion acts as a nucleophilic reagent and can be used in various organic reactions. The cyanide ion can be transformed into several other synthetic precursors, such as cyanogen bromide which acts as an electrophilic reagent to give access to an even wider variety of potential compounds.

Nitrogen-13 and Oxygen-15

N-13 and O-15 have such short half-lives that little chemistry can be done. O-15 is mainly used as O_2, water, carbon monoxide or CO_2. These radiotracers are usually prepared in flow-through systems that give very fast conversion and high efficiencies.[24]

Nitrogen-13 has been incorporated into a number of amino acids. These compounds are of interest in imaging and for the determination of protein synthesis rates in tumours.[38] A large number of amines have also been synthesised to study their distribution and function. The application of organoboranes is a very convenient method to form the amines using ammonia as the precursor. There are several other molecules, such as urea or nitrosoamines, which have been synthesised to look at various aspects of nitrogen metabolism.

Fluorine-18: Aromatic and Aliphatic Nucleophilic Reactions

In nucleophilic fluorination, F-18-labelled fluoride ion is almost always obtained as an aqueous solution. Fluoride ion is quite unreactive and requires some simple but very important manipulations to become a reactive nucleophilic reagent. A great deal of work has gone into developing methods for the preparation of reactive fluoride ion in organic solvents suitable for chemical syntheses. The steps in preparing reactive fluoride are crucial to the success of the labelling reactions and it is worthwhile to examine the methods commonly being used.

In any aqueous solution, the fluoride ion must be accompanied by positively charged counter ion. As the fluoride is removed from the target, the metal ions in the water, which were rinsed off the surface of the target during irradiation, probably serve this purpose. The reactivity of the fluoride is very effectively enhanced by the addition of a cationic counter ion prior to the evaporation of the water. Three types of counter ions have been used: large metal ions, such as rubidium or cesium; potassium complexed by a cryptand, such as Kryptofix 222;[32] or tetrabutylammonium salts.[40] Most syntheses now utilise the Kryptofix system or tetrabutylammonium salts. The addition of a cation also involves the inclusion of another anion to the reaction mixture. Anions such as hydroxide or carbonate are used, which do not effectively compete with the fluoride ion in nucleophilic displacement reactions. Carbonate is usually the anion of choice since it is less likely to cause base-catalysed side reactions.

Aliphatic Nucleophilic Displacements

The reaction of [^{18}F]fluoride ion with various leaving groups is an excellent method for the synthesis of aliphatic carbon-fluorine bonds.[41] The choice of leaving group will depend on the yield, stability of precursors, ease of subsequent separation of the [^{18}F]fluorinated product from precursors, reagents and solvents, and the formation of potential side products. Trifluoromethanesulfonate esters, commonly called triflates, are particularly reactive and provide excellent yields in nucleophilic [^{18}F]fluorination reactions, such as in the synthesis of 2-deoxy-2-[^{18}F]fluoro-D-glucose (FDG).

Halogens are good leaving groups for aliphatic nucleophilic displacements. The exchange of fluoride for chloride, bromide or iodide is quite widely used, with bromide and iodide giving higher yields than chloride. The exact reaction conditions, in terms of solvents and temperatures, are variable and must be designed for a particular reaction.

Other groups such as cyclic sulfates, mesylates and tosylates are also used as leaving groups, although they tend to give somewhat lower yields than the other groups mentioned.

Aromatic Nucleophilic Substitution

Fluorine-substituted aromatic rings are common in many types of biologically active organic molecules. Fluorine is similar in size to the hydrogen atom and does not have serious steric effects, but its high electronegativity can significantly alter the electronic characteristics of the aryl ring. Nucleophilic aromatic substitution, where F-18 is substituted for a leaving group, has become a method used widely in F-18 chemistry.[42] Nitro and trimethylammonium groups are the most widely used leaving groups in aromatic substitutions with [^{18}F]fluoride ion.[43] Simple isotopic substitution of [^{18}F]fluoride for [^{19}F]fluoride can be an effective method for synthesis of a new radiotracer. The low specific activity necessitated by these isotopic substitutions makes this reaction possible only when the specific activity of the final product is not important. Direct comparisons of nitro and trimethylammonium groups have been made, but there has been no consistent conclusion as to which gives better yields in [^{18}F]fluorination reactions. There is, however, a considerable difference in the subsequent purification of [^{18}F]fluorine-substituted aromatics. Precursors bearing the nitro-substituted aryl rings are generally carried onward in any synthesis and the final product needs to be separated from the impurity. Often, the nitro- and fluoro-substituted aromatics have remarkably similar chromatographic properties. In contrast, the trimethylammonium group is permanently charged and the precursor is chromatographically very different from the fluoroaromatic. As a result, it can be easily separated by relatively simple chromatographic methods. For nucleophilic substitution to proceed at a reasonable rate, the aromatic rings need to be activated by the presence of one or more electron-withdrawing groups positioned ortho- or para- to the leaving groups. Various substituents can function as electron-withdrawing groups, including nitro, ketones, aldehydes, nitriles, esters and amides. Studies utilising carbon-13 nuclear magnetic resonance (NMR) have shown a direct correlation between withdrawing power of a substituent and yields in nucleophilic aromatic substitutions with [^{18}F]fluoride.[43]

Electrophillic Reactions

Various electrophilic fluorinating agents have been developed for use in the synthesis of radiotracers for PET. The first to be used was F_2 derived directly from the irradiation of neon.[44] The problem was that fluorine will react with nearly everything and the highly exothermic reactions must be controlled, either with low temperature or by using very dilute solutions of fluorine in an inert solvent. The solution to this problem was to convert the fluorine into a slightly less reactive form, such as acetyl hypofluorite[45] or xenon difluoride.[46] Since all of these reagents must be prepared using carrier fluorine, the specific activity of the final product is always low. Despite these drawbacks, electrophilic fluorinations have played a vital role in the development of the radiotracers used for PET. Both FDG and Fluoro (F)-DOPA were first prepared using this method. F-DOPA is still routinely prepared with electrophilic F-18.

Clinically Useful PET Tracers

The main purpose of medical practice is to improve the outcome of the patient. This can be accomplished using

PET radiotracers designed to give information about a disease. One can use a radiotracer for preliminary diagnosis, evaluation of therapy or to determine the aggressiveness of a disease. Currently, clinical studies use FDG as the tracer. The measurement of glucose metabolic rate is one of the prime measures of physiology in the human body. PET is most often used in oncology and has demonstrated great value in this application. There are other applications as well. PET can detect viable myocardium that may respond to reperfusion. The sensitivity of PET in medically refractory epilepsy is also relatively high. In each of the following examples, PET has demonstrated improvement in diagnostic accuracy over other imaging modalities.

Oncology

In oncology, improved outcome depends, in part, on applying therapeutic strategies that are related to the stage of the disease. The importance of staging in oncology is based on the clinical experience that patients with localised malignant disease fare much better than those whose disease has spread throughout the body. Thus, staging of malignancies is essential to the selection of adequate therapy. Imaging techniques in oncology should ideally show all the detectable disease in the body. FDG-PET deserves special emphasis since it reliably meets the requirements of whole-body staging or restaging in oncology with a high diagnostic accuracy. The biochemical basis of PET cancer studies is the anaerobic glycolysis as an early indicator of malignant transformation of cells. Whole-body PET imaging with FDG enables the evaluation of glucose metabolism throughout the entire body in a single examination to improve the detection and staging of cancer, selection of therapy, and assessment of therapeutic response. Clinical indications for FDG-PET imaging are well-documented for many solid tumours in adults. An example of the uptake of FDG in a tumour is shown in Figure 3. Furthermore, encouraging early results have been published in respect of many detailed clinical questions, but confirmation in larger patient samples is being awaited. Given the clinical efficiency of FDG-PET, it is hard to justify the failure to use the technique in those patients in whom it would permit improved clinical management.

FDG-PET has been used to diagnose, stage and restage a wide variety of cancers. In lung cancer, PET gave higher sensitivity (79% vs 60%) and specificity (91% vs 77%) than CT.[47] PET is also able to scan the whole body for distant metastases. A study on colorectal cancer also showed a slightly lower sensitivity (93% vs 96%) but a much higher specificity (98% vs 69%) for PET than for CT.[47] The ability of FDG-PET to accurately stage lymphoma has been evaluated as well and shows a similar sensitivity to CT, but much better specificity (96% vs 41% for Hodgkin's lymphoma and 100% vs 67% for non-Hodgkin's lymphoma).[47] Similar results have been found with other types of cancer and in all cases, the FDG-PET techniques had higher specificity than CT.

A few other tracers have been used in oncology, such as ^{18}F-3'-fluoro-3'-deoxy-thymidine ([^{18}F]FLT) which measures cell proliferation. The first human investigation with [^{18}F]FLT was carried out in 1998.[48] The question has been whether metastases in various cancer types can be identified more accurately with [^{18}F]FLT than with the classical universal PET tool, [^{18}F]FDG.[49,50] Almost all lesions detected with FDG could be detected with [^{18}F]FLT. It has also been suggested that [^{18}F]FLT uptake is specific for malignant lesions since benign tumours do not exhibit uptake of [^{18}F]FLT.

Cardiac Studies

Imaging of the heart has proven to be extremely valuable for clinical diagnosis and an understanding of the physiology of heart in health and disease. The most widely used procedure with PET is the quantification of cardiac

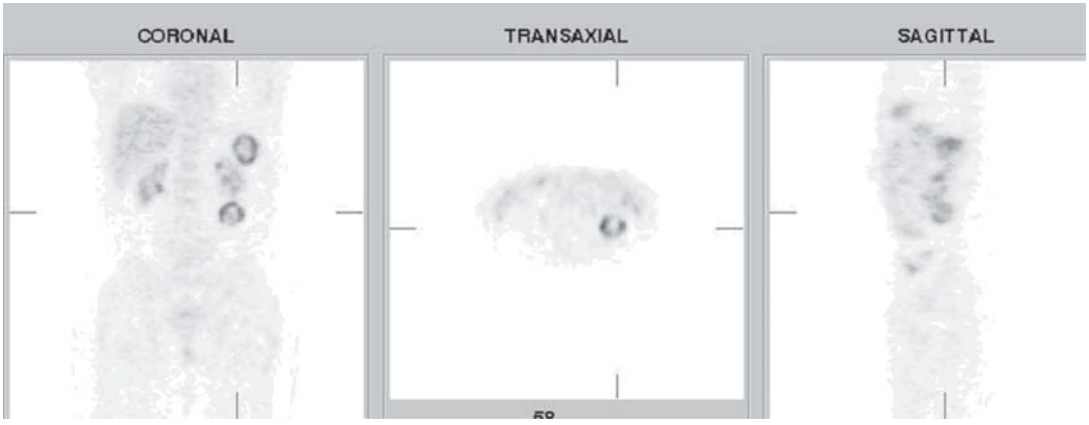

Fig. 3. Uptake of FDG in tumour located below the lung.

perfusion. This technique is used to evaluate the possibility of coronary artery disease and progress or efficacy of therapy. Metabolic imaging is carried out with FDG and this can distinguish ischaemic tissue from completely infarcted and non-viable tissue. Recently, receptor-specific radiotracers were used to evaluate the role of sympathetic, parasympathetic and muscarinic receptors, as well as how these differ in health and in disease.

Several PET tracers have been used extensively to study the heart. One of these is N-13 ammonia, which is used as a blood flow tracer in a similar manner to Tl-201 that was used in single photon nuclear medicine. O-15 water has also been used for this purpose, although not as commonly as ammonia. The myocardial oxygen consumption of the heart is often measured with C-11 labelled acetate. Again, this must be done in conjunction with a measurement of the perfusion. Fatty acid metabolism is often measured with C-11-labelled palmitate.

Tracers for Radiotherapy

Although every molecule is susceptible to damage by ionising radiation, damage to deoxy ribonuclecic acud (DNA) is the most effective in causing cell death. Whether or not tumour cells die depends on a variety of factors, including linear energy transfer, cell-cycle position, oxygen pressure (pO_2) within the tumour and the expression of a number of oncogenes and growth factors.[51] Functional imaging by PET allows the evaluation of tumour physiology, metabolism and proliferation, parameters that can determine the outcome of radiotherapy treatment.

Although numerous studies have demonstrated a relationship between the level of FDG uptake and tumour proliferation, only a few studies have addressed the predictive value of FDG uptake as an early indicator of tumour response to a therapy.[52] The results of these studies are promising and more carefully controlled studies should be completed. FDG is likely to be clinically less useful as a predictor for response to therapy in slowly proliferating tumours, such as prostate. This is because low FDG uptake values are likely to make it difficult to separate responders from non-responders. The use of radiolabelled nucleosides and amino acids is an attractive and theoretically more specific approach than FDG for visualising tumour proliferation. However, there are several drawbacks to these tracers. They are rapidly metabolised by the body, require sophisticated kinetic modelling and all use C-11 with its short half-life. To overcome these limitations, a number of halogenated analogues with sufficient half-life, which allow imaging after most labelled metabolites are washed out, have been developed and tested *in vivo* in humans, such as [^{76}Br]deoxyuridine and [^{131,124}I] deoxyuridine. Although both nucleosides are incorporated into the DNA, they are rapidly dehalogenated. This results in high background activity and low tumour uptake, thereby limiting their clinical usefulness. The relationship between tumour growth and uptake of novel molecules that are more resistant to *in vivo* degradation is an area of active investigation. As for radiolabelled amino acids, evidence that their uptake reflects tumour proliferation *in vivo* in humans has so far only been provided for [^{11}C] methylmethionine.[53] Moreover, changes in [^{11}C]methyl-methionine uptake were shown to reflect response to radiotherapy treatment in patients suffering from various tumour. The interpretation of this finding is still being speculated and more studies are warranted. Similar to radiolabelled nucleosides, the clinical value of novel radiolabelled amino acids or derivatives with longer half-life is being investigated.

Tracers for Gene Expression

The idea behind gene therapy is to use a gene to produce a missing or therapeutic protein to treat a disease or disorder. The use of a gene to supply the macromolecules overcomes the problem of delivering them through the bloodstream to a specific site. It is often difficult to determine if gene transfer is a success in patients. The definitive method is to biopsy the tissue for the gene that has been transferred (transgene), which is less than desirable. PET can be used as a tool to determine if the transfer is a success.[54-57] It can be used to either image the transgene or the expression of the gene in other endogeneous molecule.[58] Imaging transgene expression often involves a reporter gene and a reporter probe. The ideal reporter gene should generate a reporter probe only in those tissues where the transgene is expressed; if the transgene is not expressed, the reporter gene should not produce the reporter probe, and the levels of reporter probe should correlate with that of transgene expression.[59,60]

PET can be a link between biological and pharmaceutical sciences in genetically engineered and tissue- transplanted small animal models of disease and their application in the patient. The assessment of gene expression by PET is still at an early stage and has only been applied to animals. The time is soon when gene imaging by PET will play a vital role in nuclear medicine.

Outlook

PET research has been enhanced by contributions from various disciplines and has just begun to realise its potential. The number and variety of biochemical processes that can be monitored by PET are so large that only some of them have been used in clinical application. Much needs to be accomplished in the basic science of PET, particularly the radiochemistry of PET. The ability to design a molecule to assess a particular interaction is a goal for the future. Nevertheless, the ability to design a molecule that will

assess a particular interaction, and which is amenable to common synthetic routes for PET radiotracers, is a more difficult and rewarding goal.

The ability to assess the effects of gene therapy is just one of the many exciting areas that will be explored within the next few years. Much basic research needs to be done and a new generation of radiochemists are needed to develop new tracers to assess gene therapy.

One of the most exciting developments is the development of PET/CT imaging devices. These instruments combine the anatomical detail of CT with the functional information of PET in a single co-registered image that can give exquisite information relating molecular abnormalities to anatomical structures. The imaging time is also reduced because the CT data can be used to correct the PET image for photon attenuation.

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