Chapter 3
Optimization of Aseptic Production in PET Radiopharmaceuticals for Compliance to the Most Current GMP

Chih-Hao K. Kao

3.1 Introduction

Molecular imaging plays a major role in drug discovery and development because of its ability to quantify drug properties in vivo. Gradually, its potential in revealing molecular abnormalities in disease has prepared itself for an ever important application in clinical disease management. A crucial part in molecular imaging development is the scientific design and characterization of a molecular imaging agent. Among the various categories of molecular imaging agents, positron emission tomography (PET) radiopharmaceuticals are the most sensitive, which can also provide target-specific information for biochemical pathways and molecular mechanisms. For clinical consideration, PET radiopharmaceuticals do not produce detectable pharmacologic effects but provide important information concerning the etiology of various diseases. Therefore, PET radiopharmaceuticals are able to assist in the determination of optimal therapeutic dosing, delineate differential diagnosis between patients, and conceivably predict treatment responses.

These promising outlooks of PET radiopharmaceuticals result in a need for their production in industrial environments, which otherwise would not be possible to achieve in clinical settings. In keeping with this notion, manufacture of PET radiopharmaceuticals has taken steps toward mainstream pharmaceutical industry. Hence, small-scale radiolabeling performed by experienced chemists in radiochemistry laboratories or small batch production for a few patients in clinical trials is no longer a common practice. Traditional radiochemists and other PET radiopharmaceutical developers, who once found good manufacturing practice (GMP) an unfamiliar subject, were ready to equip themselves with basic indispensable concepts

C.-H.K. Kao (✉)
Buddhist Tzu Chi Foundation, Hualien Tzu Chi Hospital, Hualien, Taiwan

PET Pharm Biotech Co Ltd, Taipei, Taiwan
e-mail: chihhaokao@hotmail.com

T. Inoue et al. (eds.), *Personalized Pathway-Activated Systems Imaging in Oncology*, DOI 10.1007/978-981-10-3349-0_3

for a potential expansion of their profession into industrial PET radiopharmaceutical production. While terminologies such as quality assurance programs, qualification and validation, and environmental monitoring are no longer foreign, there are still significant challenges ahead before a fully integrated "PET drug production" is able to claim itself readily compliant to the most current GMP standards. Some of the challenges were too easily deemed "impossible to overcome" or "not applicable" for PET radiopharmaceutical before today's production. However, they were in fact norms to all other types of pharmaceutical production and thus "nonnegotiable" in the eyes of GMP inspectors. Can we still use "small batch volume" and "short half-life" as excuses to circumvent these issues? More often than not, a solution was just not thought out hard enough, and ingenious techniques were eventually created to resolve the problem only upon GMP inspector's insistence. Nevertheless, PET drug production is progressing and getting its acceptance into pharmaceutical industry. This chapter describes a part of this endeavor that is the most unfamiliar to traditional PET radiopharmaceutical production and provides a few examples on how these could be optimized.

3.2 Trends in PET Radiopharmaceutical Development

For decades, development of PET radiopharmaceuticals centered on molecules labeled with fluorine-18 (^{18}F, half-life, 110 min), carbon-11 (^{11}C, half-life, 20 min), and a few other halogen isotopes with much longer half-lives such as bromine-76 and iodine-124. These radionuclides are cyclotron produced and are incorporated into biochemically active molecules via covalent chemistry. Fluorine-18 radiopharmaceuticals in particular have prevailed due to, among other factors, its more appropriate half-life. For instance, the technology for producing ^{18}F-fluorodeoxyglucose (FDG) is now so mature that a large quantity of FDG can be obtained by touches of a few buttons, and its quality as a medicinal product meets the highest standard for a sterile injectable solution. FDG is currently the single most widely used PET radiopharmaceutical, which has been so successful in imaging disease with high glycolytic rate and, thus, has become one indispensable tool in the management of many cancers [1]. However, FDG has several limitations, which can give rise to false-positive/false-negative diagnoses and poor predictive value of tumor chemoradiation therapy responses. In addition, certain tumors such as the neuroendocrine type have poor uptake of FDG [2]. Therefore, there is a high demand to develop new radiopharmaceuticals beyond FDG in the oncology field, and indeed many are already in various stages of development. One important feature that sets them apart from FDG and many other PET radiopharmaceuticals of the earlier days is not scientific but methodological in terms of how they were developed. Most of these new and promising PET radiopharmaceuticals were being moved forward in industrial

settings, and their synthesis methods were chosen based on automation capabilities and yields. A pharmacologically favorable PET radiopharmaceutical might never make its way into clinical medicine if a commercially viable production method could not be found.

A similar argument can be made for PET radiopharmaceuticals in the neurological field. In fact, the first new PET radiopharmaceuticals in decades to be granted marketing authorizations in major countries and regions such North America and Europe are those designed for the detection of abnormal protein plaque deposition in the brain [3].

A bright prospect in new PET radiopharmaceuticals is the development of gallium-68 (^{68}Ga, half-life, 68 min) compounds, which represent an entirely different category, in terms of their chemistry, their production, and supply routine. The generator-produced metallic radionuclide ^{68}Ga is conjugated to a small peptide via coordinating ligands and serves as a convenient alternative to ^{18}F for routine clinical practice. A number of ^{68}Ga-labeled peptides have proven their efficacies in imaging neuroendocrine tumors and prostate cancers [4].

3.3 Regulations and Standards for PET Radiopharmaceutical Manufacture

Regardless of ^{18}F or ^{68}Ga compounds, these short-lived radiopharmaceuticals are characteristically different from nonradioactive drugs and even from other longer-lived radiopharmaceuticals such as iodine-131 capsules and technetium-99 m compounded solutions. Nevertheless, they are medicinal products for human use, and, hence, their manufacture must be subjected to proper regulations in order to control their quality. Moreover, because PET radiopharmaceuticals are virtually always solutions for intravenous injection, extremely good practice manufacture has to be guaranteed, to eliminate the risk of product contamination. Since they are usually not able to wait through a longer process of terminal sterilization to completely kill potential contaminating pathogens, aseptic handling procedures are critical during production. Collectively, all these necessary steps make the manufacture practice of short-lived PET radiopharmaceuticals one of the toughest GMP processes to follow and comply.

An official GMP guidance is usually issued and enforced by individual national governments, and it generally covers the same manufacturing process and facility, quality guidelines, and personnel training. However, some standards and thus the quality of products may vary to an unacceptable degree. To solve this problem, the Pharmaceutical Inspection Convention (PIC) and the Pharmaceutical Inspection Co-operation Scheme (PICS) have created a universally concurred GMP guide for medicinal products. PIC and PICS (PIC/S) are two international instruments, which

jointly provide an active and constructive cooperation in the field of GMP, between countries and pharmaceutical inspection authorities. Their mission is to lead the international development, implementation, and maintenance of harmonized GMP standards and quality systems of inspectorates in the field of medicinal products (http://picscheme.org/).

With obvious distinct characteristics, radiopharmaceutical production was given a dedicated section, Annex 3, in PIC/S GMP guide. Annex 3 covers all radiopharmaceuticals while offering some exemptions for short-lived PET radiopharmaceuticals. The exclusion of the cyclotron portion in the production section of this guide means that "GMP" starts from the point in which the radioactive starting material (like a solution containing ^{18}F) enters into the production process. Additionally, a very rare process of "conditional release" of final product is allowed so that a short-lived product can be released for patient use, before completion of the sterility test. However, being an aseptically processed product, short-lived PET radiopharmaceuticals are subjected to one of the most difficult sections of all: Annex 1, for sterile medicinal product, which makes this PIC/S GMP regulation very difficult to comply.

In the United States, a version of GMP is specifically designed for PET radiopharmaceuticals in Section CFR 21 part 212, which is a cGMP rule for PET drug production. This US version is generally considered easier to comply because it acknowledges that a short-lived PET drug is in fact "short-lived" which implies that the potential harm a contaminated product can cause is considerably limited. The risk it carries and thus the rule for its manufacture should not be comparable to those of just any other sterile medicinal products.

3.4 Example of a Cleanroom Designed Specifically for Aseptic Production of PET Radiopharmaceuticals

The PIC/S GMP guide specifies cleanroom grading with Grade A, as being the cleanest, versus Grade D, designated as the least clean. Table 3.1 lists the limits of particle number permitted in each clean grade. This guide also specifies grading requirements for various types of activities to be conducted. For example, an automatic synthesis module needs only be placed in a Grade C room, but the final filling of a filtered solution needs to be done in a Grade A space, with a Grade B background. The design of a cleanliness-controlled complex for a PET radiopharmaceutical production laboratory (lab) should follow those guides.

A conventional PET radiopharmaceutical production room in a university lab or research hospital usually has multiple hotcells enclosing various synthesis modules. Each module is connected by a dedicated fluid transfer tube that transports a formu-

Table 3.1 Maximum permitted number of particles/m^3 equal to or greater than the tabulated size

	At rest		In operation	
Grade	0.5 pe	5.5 m	0.5 pe	5.5
A	3520	20	3520	20
B	3520	29	352,000	2900
C	352,000	2900	3,520,000	29,000
D	3,520,000	29,000	Not defined	Not defined

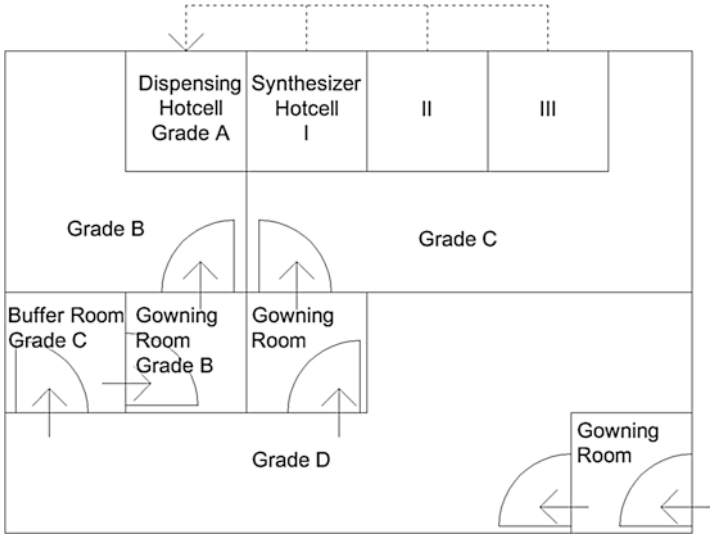

Fig. 3.1 This is a conceptual drawing of a conventional PET radiopharmaceutical production complex that follows basic cleanroom requirement of PIC/S GMP guidance. A row of three hotcells for synthesizers is in a Grade C room, while a Grade A dispensing hotcell is in a separate Grade B room. *Dotted lines* indicate transport tubes from each synthesizer. This drawing does not depict necessary functions such as pass boxes for material in and product out

lated product to a distant hotcell for dispensing and packaging [5, 6]. This dispensing space is responsible for handling multiple incoming formulated products from different synthesizers. Aseptic filtration of formulated products is usually performed in this hotcell as well. This type of space is not too difficult to be transformed into a cleanliness-controlled complex according to the above guidance, as long as the dispensing hotcell can be kept in a Grade A room, which is located at Grade B environment, although this is only from the viewpoint of cleanroom grading (Fig. 3.1). There are many other designs that might better suit various purposes in PET radiopharmaceutical production. As an example, this section describes a new concept in an industrial PET radiopharmaceutical manufacture facility. This site is a centralized PET radiopharmaceutical provider in Taiwan, and it has been certified according to PIC/S GMP guide by the Taiwanese authority (Fig. 3.2).

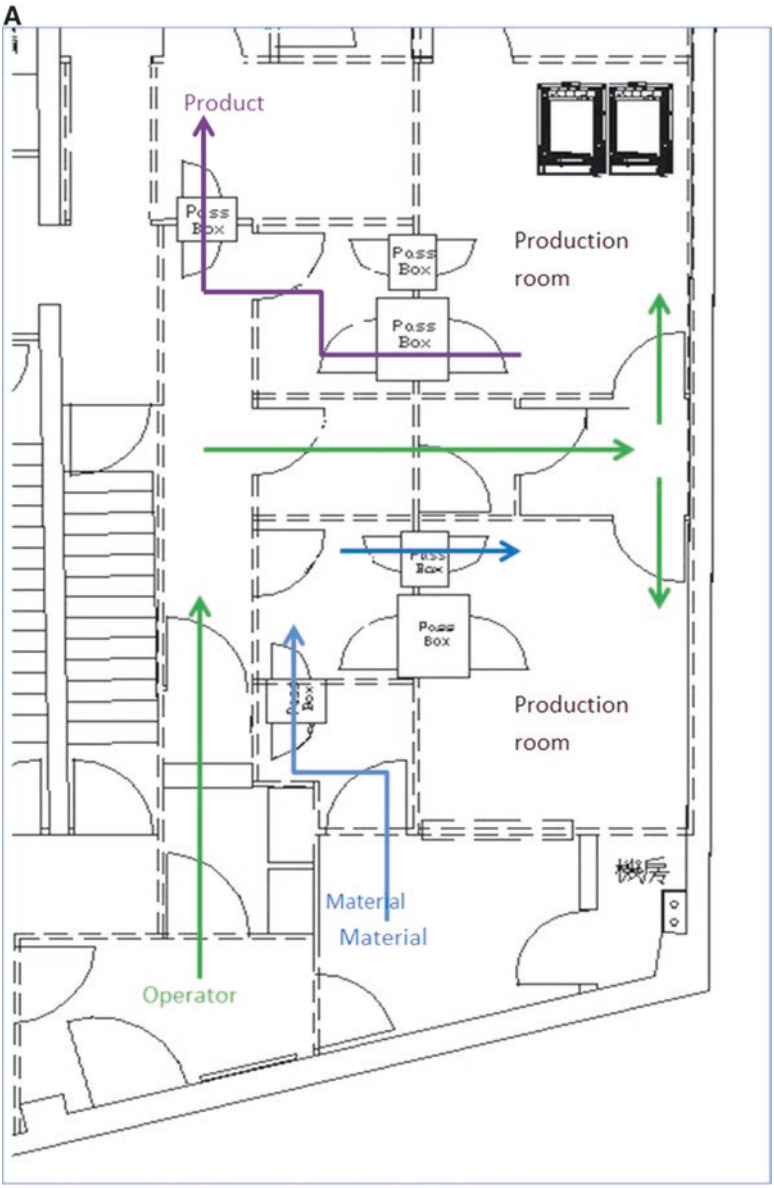

Fig. 3.2 This is the actual cleanroom complex in PET Pharm Biotech Co Ltd. (**a**) Floor plan. Operators enter through *left bottom* corner (*green arrow*) into a Grade D corridor. The two production rooms are assessed via series of small buffer spaces. Only the upper production room is depicted with a hotcell in this drawing. (**b**) 3D rendition of the floor viewing from the *left side* of the plan. *Crimson* color floor is Grade D while *gray* C and *yellow* B. The final gowning and two production rooms are in Grade B. Two rooms with pass boxes in Grade C are for passing of materials and products. (**c**) Viewing from the opposite angle

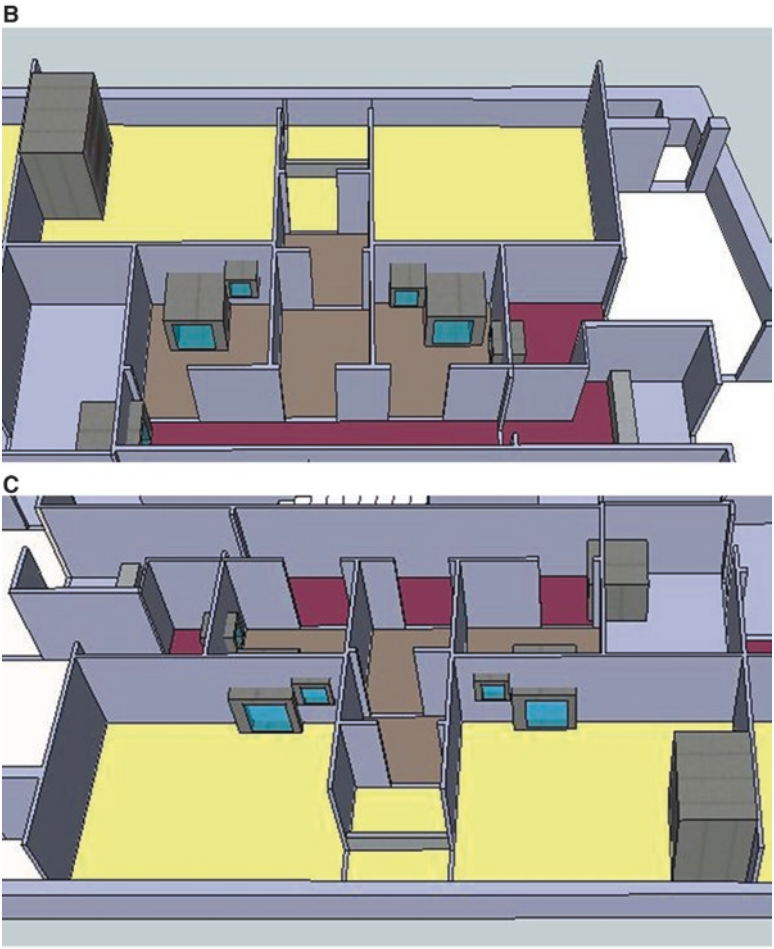

Fig. 3.2 (continued)

This alternative design argued that multiple small Grade B rooms, each setup with a hotcell encasing one Grade B synthesis compartment and one Grade A aseptic filling compartment, would be a more efficient way for complying the toughest GMP, especially in a site that more than one kind of PET radiopharmaceuticals are regularly produced and dispensed.

Entering via a series of small rooms that increase in the cleanliness grades, the Grade B final gowning room opens to two Grade B production rooms. In each production room (12 m^2) stands one hotcell with two main compartments. The synthesis compartment is not controlled for its air particles to a specific standard, but because it opens to the production room, and thus it is in the same level of cleanliness as the production room itself. The other compartment of the hotcell is for asep-

tic filling; it is therefore designed to be an isolator of Grade A. Inside this isolator, particle counts are monitored continuously and air is sampled for bacteria.

The concerned cleanroom complex is only 38 m^2 including buffer, final gowning, raw materials and products in and out, and two production rooms. There is a system established to monitor temperature, humidity, radiation dose rate, and air pressure in each room. The ventilation control system has chambers to collect waste gas as well as to vent through hotcell. With each production room only 12 m^2, one hotcell, and maximum two operators, Grade B is easily maintained. The regular and frequent disinfection practice required for the Grade B room is also achieved without difficulty. Within hotcell the two compartments are adjacent, and thus the product solution transport line is short enough to allow the use of the prepackaged disposable sterilized tubing. More importantly, two production rooms can be in operation simultaneously with diminished risk of cross contamination. The government auditors have deemed this design in compliance with PIC/S GMP requirements.

Cross contamination among different drug products is one of the major issues that PIC/S GMP requirements designed to avoid. Yet, in an effort to comply with these rules, most PET radiopharmaceutical manufacture facilities still set up their production rooms with multiple hotcells, so that various PET radiopharmaceuticals could be produced on the same day even simultaneously. To make the matters worse, these different products often share the same aseptic filling space. Though the synthesizer room is only required to maintain a Grade C cleanliness, the Grade A aseptic filling still needs to be stationed in a Grade B environment, which requires a preceding Grade B gowning room as depicted in Fig. 3.1. The overall use of space is not as economical as one might think, and it would be hard to convince GMP auditors that cross contamination would be unlikely.

This “one room, one product” concept simplifies the work flow and allows two rooms in operation simultaneously. No different batches of PET radiopharmaceuticals will be processed in the same hotcell space, which greatly lowers the risk of cross contamination. Though now the synthesis module is set up in Grade B environment, this only enhances the quality of production. In conclusion, this cleanroom design is effective and in compliance with the most current and toughest GMP requirements.

3.5 Challenges in Aseptic Process in Short-Lived PET Radiopharmaceutical Manufacture

Short-lived PET radiopharmaceuticals such as FDG are usually produced as a batch of solution in small volume. The volume of the original solution coming out of a dedicated FDG synthesizer can be as small as 12 mL, but it is usually around 20 mL. The product solution is filtered to achieve sterility by flowing from the synthesizer through a filter-connected tube into a batch vial. After its final closure, the vial is properly packaged and sent out as a final product to a radiopharmacy, where it is

subsequently dispensed into a unit-dose syringe for each individual patient. This final product package is called a multidose vial because it usually contains more than enough FDG for a single-patient dose. Alternatively, one can dispense into multiple unit-dose syringes filtered product solution directly. A filled syringe is then the product's final package. Irrespective of what final product packaging is chosen for one manufacture site, this process has to be performed in Grade A compartment and handled aseptically, which means following the Annex 1 of the PIC/S guide in all details.

In this Grade A hotcell compartment that is validated for HEPA functions, airflow speed and pattern, air pressure differential, temperature, and humidity are merely basic requirements. Aseptic processing for manufacturing a sterile product has many other obligatory functions that demand real challenges.

Design Qualification (DQ) for a Dispensing Hotcell DQ is a process to be done prior to the better known 3Q (IQ, OQ, and PQ). It is actually a requirement in many parts of the GMP but quite often neglected. One, however, would find oneself feeling stranded if DQ was neglected for a dispensing hotcell. Here are non-exhaustive examples explaining this situation. A hotcell needs to have certain built-in features in order to accommodate certain obligatory functions, which in turn show how important the DQ is for this hotcell:

1. Continuous monitoring of particle counts during aseptic processing is nonnegotiable and thus needs to be done. A built-in particle counter in the hotcell certainly fulfills the demand, but an off-the-self hotcell is not likely to have this feature. Alternatively, a commercial particle counter with a long particle-collecting tube might also serve the purpose, if only the tubing could penetrate through the hotcell wall.
2. Continuous air sampling is another similar situation because air exhaust from the sampler needs to go outside the hotcell.
3. Fumigation by disinfectant such as hydrogen peroxide before each production, which usually means at least daily for PET radiopharmaceutical, is another requirement. A normal hotcell interior material and fixtures usually do not expect an environment as harsh as this, and things break down much easier than usual.

Sampling for Bioburden Analysis "Bioburden" is a term that describes microbiological burden brought by an unsterilized product. To analyze this is to understand sources of microbiological contamination for a specific production process. In order to do this, one must take a sample from the product before it is being subjected to sterilization and microbiological analysis, which is subsequently tested. This procedure must be performed for each and every product batch produced.

PET radiopharmaceuticals such as FDG are usually produced by an automatic module, and the process includes radiochemical reactions and product purification and formulation, which are performed in a close tube system all the way to sterile filtration and until the product solution is dispensed into final the container. Now in order to take a sample for bioburden test, this "nonstop and all-the-way flow through

in a close tube" system could suddenly become impossible. More difficult still is the requirement of minimum volume sampling that usually is not less than 10 mL, whereas the batch of product might only be 20 mL at best. Achieving such task is not trivial and it will really put one's ingenuity to the test. It is not practical to provide any answer here as manufacture sites vary in their synthesis module, filtration apparatus, and many other configurations. Yet readers need to consider these issues in the design of their own production flow.

3.6 Conclusion

The design of most current GMP facilities for PET radiopharmaceutical manufacture around the world has gradually gained momentum during the past decade. While basic requirements in premises, equipment, personnel, documentation, and operations are no longer headaches to some, details abound with difficult issues for most involved. Perhaps, regulatory auditors may finally yield on a few things based on low-risk factors. This chapter merely initiates the process by pointing out challenges by presenting some examples. It is the management and operators everywhere who would eventually provide specific solutions to optimize the respective facilities.

References

1. Delbeke D, Coleman RE, Guiberteauetal MJ. Procedure guideline for tumor imaging with ^{18}F-FDG PET/CT 1.0. J Nucl Med. 2006;47(5):885–95.
2. Eriksson B, Bergström M, Orlefors H, Sundin A, Oberg K, Långström B. Use of PET in neuroendocrine tumors. In vivo applications and in vitro studies. Q J Nucl Med. 2000;44(1):68–76.
3. Sabri O, Sabbagh MN, Seibyl J, Barthel H, Akatsu H, Ouchi Y, et al. Florbetaben PET imaging to detect amyloid beta plaques in Alzheimer's disease: phase 3 study. Alzheimers Dement. 2015;11(8):964–74.
4. Sharma P, Singh H, Bal C, Kumar R. PET/CT imaging of neuroendocrine tumors with ^{68}Gallium-labeled somatostatin analogues: an overview and single institutional experience from India. Indian J Nucl Med. 2014;29(1):2–12.
5. Kao CHK, Hsu WL, Xie HL, Lin MC, Lan WC, Chao HY. GMP production of [18F]FDOPA and issues concerning its quality analyses as in USP "Fluorodopa F 18 Injection". Ann Nucl Med. 2011;25(5):309–16.
6. Kao CHK, Hsu WL, Kao PF, Lan WC, Xie HL, Lin MC, et al. An efficient and aseptic preparation of "sodium fluoride (18F) injection" in a GMP compliant facility. Ann Nucl Med. 2010;24(3):149–55.