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Synthese neuartiger Adenosin-A₃-Rezeptor-
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1. INTRODUCTION

1.1 Basics

1.1.1 Adenosine^{1, 2, 3}

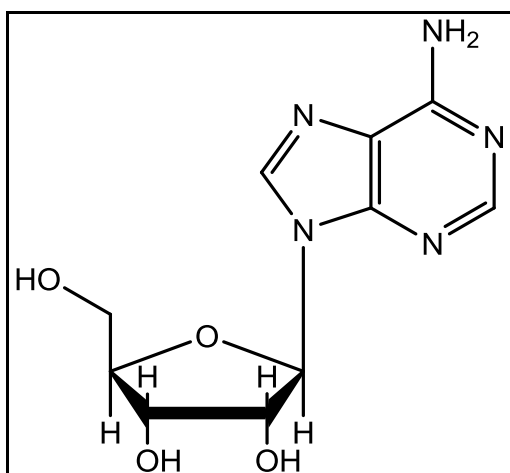


Fig.1 Adenosine

Adenosine is an endogenous key molecule, consisting of the purine base adenine, linked to the monosaccharide β -D-ribose. Adenosine is omnipresent in all species and plays a pivotal role in numerous biochemical processes such as signal transduction pathways as cyclic adenosine monophosphate (cAMP), as inhibitory neurotransmitter in the central nervous system and as energy carrier necessary to sustain living systems.

Adenosine inhibits the release of excitatory and activating neurotransmitters like dopamine, acetylcholine, or noradrenaline, causing vasodilatation, thus reducing blood pressure. Furthermore, adenosine decreases heart frequency and prolongs conduction time (PQ) in the sinoatrial node, making it a diagnostic drug for testing and/or treating different types of supraventricular tachycardia.

Adenosine has also sleep inducing properties by activating the ventrolateral preoptic nucleus (VLPO), a group of neurons in the hypothalamus releasing the inhibitory neurotransmitters galanin and GABA as well as inhibiting other neurons involved in wakefulness.

Adenosine can be attached to one, two or three phosphate groups building adenosine monophosphate (AMP), adenosine diphosphate (ADP) or adenosine triphosphate (ATP). ATP is an energy rich molecule due to its two reactive phosphoanhydride bonds of the triphosphate unit. When hydrolyzed, large amounts of free energy are liberated, used for almost all energy-consuming processes like motion, active transport, or biosynthesis.

1.1.2 Adenosine receptor types^{1, 2, 4, 5, 16}

To date, four different adenosine receptor subtypes have been identified in several mammal species, including man. All of them are G-protein-coupled receptors, consisting of seven transmembrane domains. They are called P₁-purino receptor and are distinguished from the P₂-purino receptors that bind the nucleotides ADP and ATP.

The A₁ receptor subtype is expressed in the entire central nervous system and shows highest levels in the cerebral cortex, hippocampus, cerebellum, thalamus, and brain stem activating K⁺ channels and inhibiting Ca²⁺ channels - both of which reduce neuronal activity.^{25, 26}

The A_{2A} receptor subtype is expressed only in few regions of the brain at high levels, namely the dorsal and ventral parts of the striatum^{17, 18} (Nucleus caudatus, Putamen and Nucleus accumbens) activating adenylyl cyclase.

The A_{2B} receptor is ubiquitous in the brain²⁷, which makes it difficult to connect it to specific physiological and behavioural properties, because there are only few known specific A_{2B} agonists and antagonists. The A_{2B} receptor activates adenylyl cyclase.

The A₃ receptor subtype is little investigated, however it is believed to uncouple A₁ and metabotropic glutamate receptors via protein kinase C, thus modulating activity of other receptors.

Recent studies showed that the adenosine A₃ receptor is expressed in almost all kinds of tissue like brain, heart, liver, kidney, or intestines.

Since most studies are based on mere mRNA quantification, only little information regarding absolute receptor distribution and density is available.¹⁴ In the attempt of developing new techniques of quantification of the adenosine A₃ receptor, positron emission tomography (PET) proved to be the most suitable method, although suitable radioligands were required to perform this task.

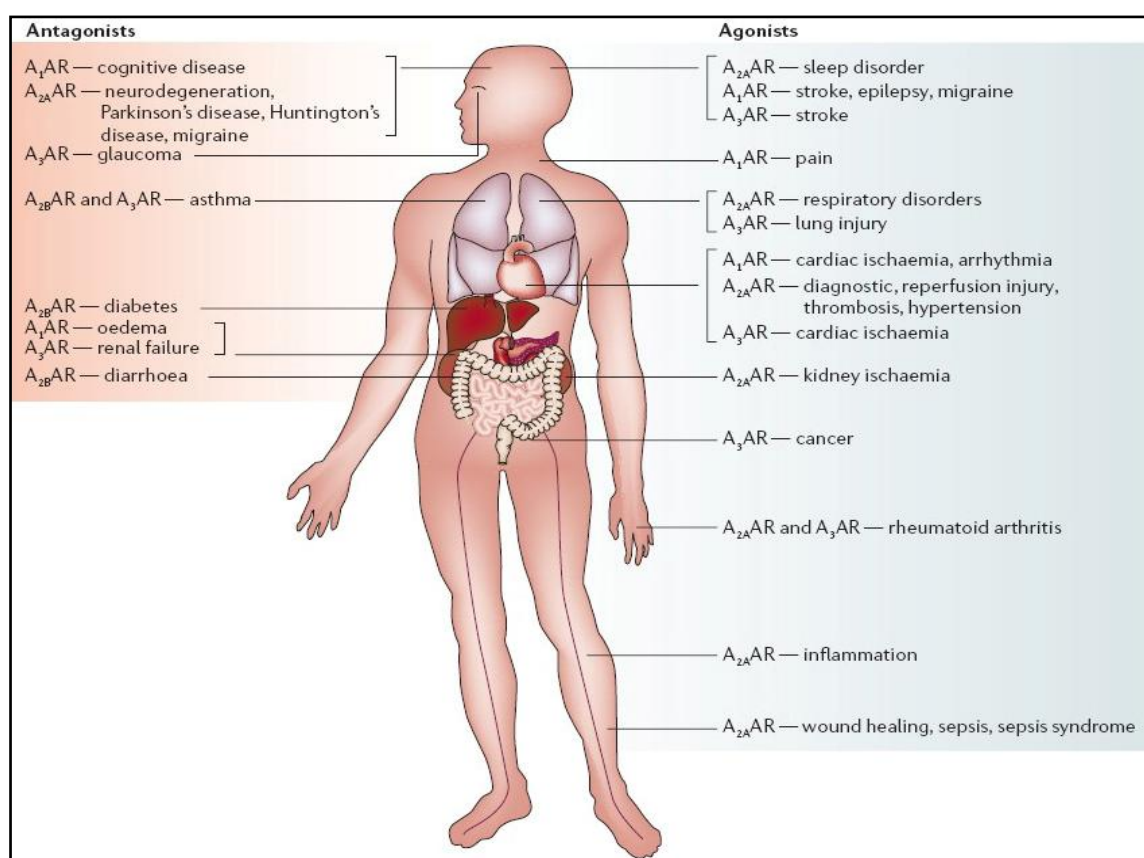


Fig.2 Possible drug targets for adenosine receptor subtypes⁵

1.1.3 The adenosine A₃ receptor and pathological conditions^{1, 2, 16}

The adenosine A₃ receptor is present in almost all kinds of tissue associated with many diseases like heart⁷ and brain ischaemia⁸, glaucoma⁹, stroke¹⁰ and epilepsy¹¹. It is also involved in chronic inflammatory processes, asthma, and cancer¹². Recent studies showed the A₃ receptor is expressed at high levels on the surface of

primary and metastatic tumour cells^{19, 20, 21, 22, 23, 24}, making it a promising target for future anti tumour compounds as well as tumour imaging techniques.¹³

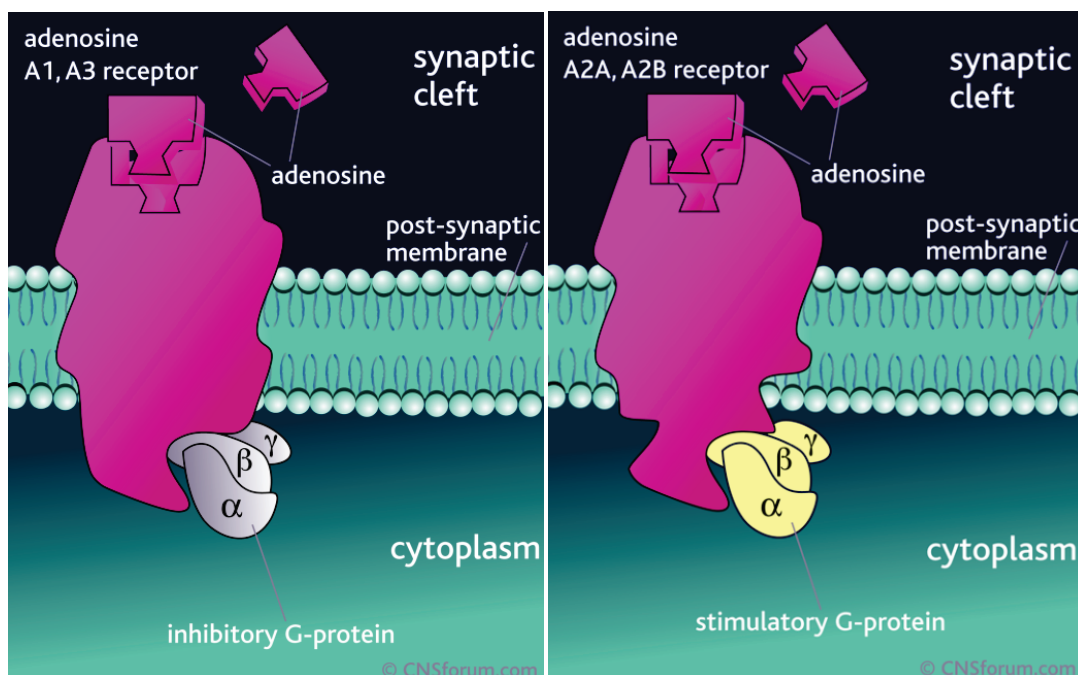


Fig.3. Schematic of the adenosine receptor types A₁, A_{2A}, A_{2B}, and A₃¹⁵

1.2 Radioactive Decay^{28, 29}

Radioactivity is the ability of an unstable atomic nucleus to lose energy by emitting ionizing particles or gamma radiation. This results in the transmutation of the so-called parent nuclide into another chemical element, called the daughter nuclide. The process of radioactive decay is spontaneous and random, which means predicting the decay of a certain atom is not possible, but in a large number of given atoms it is possible to predict the rate of decay in average over all atoms. The probability of radioactive decay is expressed by the half-life, defined as the time in which 50 percent of a given number of unstable nuclides have undergone radioactive decay. Half-life is constant and characteristic for each unstable element. Half-lives vary widely, from 10^{-23} seconds for highly unstable nuclides to 10^{19} years for ^{209}Bi .

The SI unit is Becquerel (Bq), which is defined as one decay per second, but usually Gigabecquerel (GBq) is used as to the large number of atoms in regular samples of radioactive material.

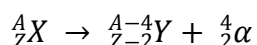
The cause for the instability of some elements lies in the composition of the nucleus. A nucleus is composed of neutrons and protons that are controlled by the strong nuclear force, the electrostatic force and, in case of beta decay, by the weak force. If the balance between them is disturbed, the nucleus releases its energy either as gamma radiation photon or as particle like an alpha particle or a positron. This transformation alters the structure of the nucleus, yielding an element of higher stability.

1.2.1 Modes of decay^{28,29,30, 32, 33, 34, 35}

Dependent of the composition of the radionuclide, different modes of decay are possible. We distinguish between decay emitting nucleons, like alpha decay, the emission of an electron or positron called beta decay and the emission of high energy photon emission with no matter emitted at all, called gamma decay.

1.2.1.1 Alpha decay³²

Alpha decay is the type of decay in which an atomic nucleus ejects an alpha particle, which is a helium nucleus. In this process, an atom with a mass number (A) minus 4 and atomic number (Z) minus 2 is formed as shown below.



The helium nucleus is a doubly charged helium atom; therefore, alpha radiation is a form of ionizing particle radiation. The alpha particle has a typical kinetic energy of 5 MeV and leaves the nucleus at a speed of approximately 15, 000 - 20, 000 kph. The remaining nucleus sometimes remains in an excited state emitting gamma photons in order to remove the excess energy.

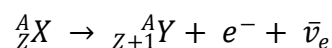
Alpha decay usually occurs in heavy elements like uranium, thorium, radium, and the transuranium elements and must have a minimum-size atomic nucleus. The lightest elements ever observed to emit alpha radiation are the nuclides of tellurium with mass numbers of 106 to 100.

As for their large mass and low velocity alpha particles interact with other atoms, eventually losing their energy – which is the reason why they can be easily shielded by a sheet of paper or a few centimetres of air. Although alpha radiation is unable to penetrate skin and therefore is relatively harmless when exposed from outside, inhaling or by food intake can cause severe damage to living cells. Due to its strong ionizing properties and long half-life, this type of radiation is unsuitable for diagnostic purposes.

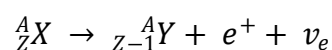
1.2.1.2 Beta decay^{33, 35}

Beta radiation is, similar to alpha decay a type of high-energy ionizing radiation, caused by electrons or positrons emitted by radioactive nuclei with an excess of neutrons or protons. If an electron is emitted it is referred to as beta minus (β^-), while in case of a positron emission as beta plus (β^+). Unlike in alpha decay, beta decay is neither governed by nuclear nor electromagnetic force, but by weak interaction.

In beta minus (β^-) decay, a neutron is converted into a proton, while emitting an electron (e^-) and an electron antineutrino ($\bar{\nu}_e$). An atom with the same mass number (A) and atomic number (Z) plus 1 is formed as shown below.



In beta plus (β^+) decay, a proton is converted into a neutron, a positron (e^+) and a neutrino (ν_e). An atom with the same mass number (A) and atomic number (Z) minus 1 is formed:



The emitted positron is not very stable when in contact with matter, leading to an annihilation event once the positron collides with an electron. In this process, two gamma ray photons of 511 keV are being produced and emitted in opposite

direction. These two gamma photons are used for positron emission tomography (PET).

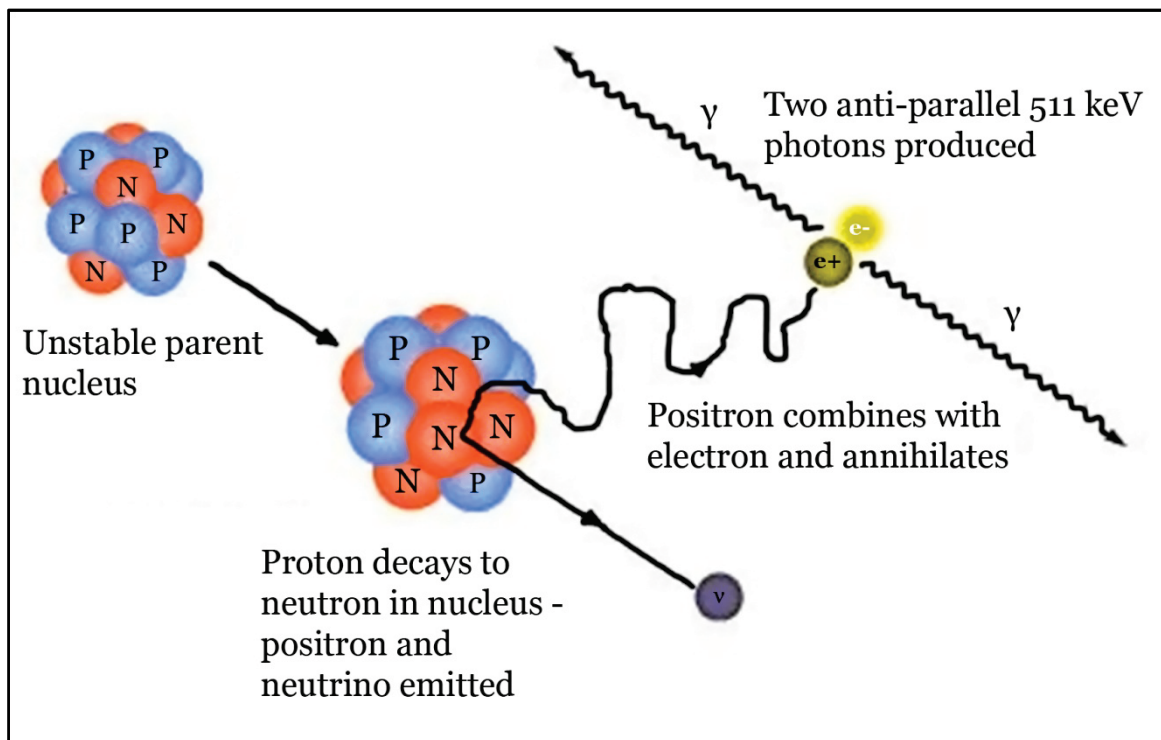
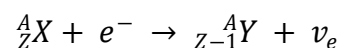


Fig.4 Annihilation (modified)³⁶

A related mode of decay is the electron capture which, like beta plus decay, occurs in a nucleus containing too many protons, but unlike in beta plus decay, the energy necessary to emit a positron is insufficient - that is an energy difference of parent and daughter nuclide less than 1.022 MeV.

Instead, an orbital electron is captured by a proton and converted to a neutron and a neutrino (ν_e). Since the proton is changed to a neutron, the mass number (A) remains unchanged and the atomic number (Z) is decreased by 1, transforming the nuclide into a new element:



If the resulting kinetic energy is not transferred entirely to the neutrino, the newly created nuclide remains in an excited state. When transiting to the ground state, a gamma photon, and/or Auger electron is emitted.

Beta radiation has more penetrating powers than alpha radiation, but is far less penetrating than gamma radiation, although its ionising powers are stronger due to its composition of charged particles. The energy released by beta particles varies, but the typical maximum energy is approximately 1 MeV. Nevertheless, they can be stopped by a few millimetres of aluminium. Passing through matter decelerates beta particles by electromagnetic interactions, which may cause bremsstrahlung gamma radiation.

Beta particles penetrate the skin and can severely damage the skin layers, causing burn and long-term effects such as cancer. If beta particles are taken up internally, high radiation contamination in the surrounding tissue can occur, which has been proven for thyroid cancer as long time effect from ^{131}I exposure. Exposing of the eyes to beta radiation can cause cataract. The accumulation of radioactive ^{90}Sr in the skeleton is believed to cause bone cancer and leukaemia.

Ironically, beta particles are used in radiation therapy to treat several kinds of cancer as to their ability to kill cancer cells as well.

Another field of application is medical imaging, in which certain tracer molecules are used as source for positrons yielding from beta plus (β^+) decay.

1.2.1.3 Gamma radiation³⁴

Gamma radiation is electromagnetic radiation of high frequency and short wavelength. It is caused by subatomic particle interactions such as electron-positron annihilation, neutral pion decay, radioactive decay, fission, fusion, or inverse Compton scattering. Typical gamma ray frequency is above 10^{19} Hz; therefore, the wavelengths are less than 10 picometres and energies above 100 keV. Gamma rays caused by radioactive decay usually have energies between a few 100 keV and less than 10 MeV. Gamma radiation is ionizing radiation, such as alpha or beta radiation, but of more penetrating powers.

In the past, gamma rays were distinguished from X-rays based on energy amounts, considering gamma rays as more powerful than X-rays. Nevertheless, as energy ranges overlap widely, this distinction became obsolete. The two are the same in essence, the only difference between them being their creation process.

Gamma rays are caused by nuclear decay, whereas X-rays are emitted by electrons in orbitals outside the nucleus or when accelerated to produce bremsstrahlung radiation.

Gamma radiation is the most penetration radiation, requiring heavy shielding³¹ to be protected against it. Large amounts of mass are necessary. Elements with high density and high atomic number are ideal. The probability for absorption is proportional to the thickness of the shielding, leading to an exponential decrease of radiation intensity with thickness. Lead is a very dense metal, yet cheap and compact, making it adequate for that purpose. Concrete is more suitable for large radiation sources and depleted uranium is applied in portable gamma ray sources in order to reduce bulkiness.

Gamma radiation, like any ionizing radiation, is harmful for organic life, causing damage at a cellular level. Alpha and beta radiation, however, are less dangerous, because they cannot penetrate the body easily and mostly cause localized damage like radiation burn to the skin. Gamma radiation penetrates the body easily, therefore causes damage to the DNA. Subsequently, this leads to interference with the cell's ability to divide and duplicate, rather than instantaneous cellular breakdown. This effect can be observed in fast proliferating cells such as enterocytes, hair follicles, or skin cells within a short period. Cellular necrosis occurs, ranging from minutes to days, depending on the level of exposure, whereas in slowly proliferating cells this effect might be observed years later. The reason for this is the inability of the remaining stem cells to duplicate; therefore, this effect is immediate in cells with fast division cycle and late in slowly duplicating cells. These symptoms are called radiation sickness or radiation poisoning.

Despite its devastating effect on living tissue, gamma radiation of all types of radiation has the widest field of application. It is used for sterilizing single-use medical equipment such as syringes or needles or sterilizing food cans, chemical sciences make use of gamma radiation in x-ray-crystallography for structural determination of many inorganic and organic molecules including biopolymers such as proteins. It is also used in industrial radiography as material testing method for structural damage, debris, or corrosion. Gamma ray detectors are used at airports to screen people and luggage and at harbours to screen ship containers. Its medical application includes many imaging diagnostic techniques like x-ray radiography, computed tomography (CT), single photon emission computed

tomography (SPECT), as well as means of treatment for cancer including gamma-knife surgery and brachytherapy.

1.2.2 Natural radiation³⁷

Background radiation is omnipresent in our environment, emitted from natural sources and artificially created ones. Natural sources mainly have two origins, cosmic radiation, and terrestrial sources.

Radiation from outer space is generated, when positively charged ions from outside the solar system hit the atmosphere. Secondary radiation, including gamma radiation, protons, alpha particles, electrons and neutrons, is created. The dose varies in different parts of the planet, depending on the altitude and the geomagnetic field. In the upper troposphere at around 10 kilometres, the altitude where passenger aircrafts travel, the radiation is more intense, posing a certain danger for long-distance flight crews and frequent passengers.

Cosmic radiation also causes elemental transmutation by interaction with atomic nuclei in the atmosphere. The most relevant are the ^{14}C isotopes, which are produced by interaction with nitrogen atoms. Eventually, they reach the earth surface and become incorporated to living organisms. This is the foundation of radiocarbon dating.

On earth, radioactive isotopes are present in soil, rocks, water, air, and vegetation and were formed billion years ago by stellar nucleosynthesis. These radioactive isotopes can be from common elements like potassium (^{40}K) or carbon (^{14}C) or the long-lived elements such as uranium (^{238}U) or thorium (^{232}Th) and their decay products like radium or radon. The level of radiation varies depending on the area. Some locations have significantly higher background radiation like Kerala in India, Yangjiang in China or Ramsar in Iran. The latter has up to 200 times greater background radiation than in average, due to hot springs in the surrounding area that are used as spas. Nevertheless, there seem to be no ill effects on the residents associated with the excessive radiation; on the contrary, the locals seem to be slightly more radio resistant. Based on this „radiation paradox“, some scientist suggest that the „linear no-threshold model“, on which

all radiation regulations are based, is not valid and that a certain amount of radiation might be beneficial for health.^{51, 52}

1.2.3 Artificial radiation^{37, 38}

All kinds of artificial radiation are man-made radiation and identical in their nature and their implications and add to the natural occurring radiation. Sources are nuclear facilities like nuclear power plants (especially the Chernobyl incident), medical imaging, and radiotherapy, fallout from nuclear weaponry testing and research facilities. The creation of radioactive isotopes is accomplished by bombardment of a nucleus with alpha particles, protons, neutrons, or gamma radiation photons, leading to the artificial transmutation. The created radionuclides do not exist on earth naturally and, as result of their short half-lives, have to be created in-situ for each application purpose. E. Rutherford created the first artificial nuclei in 1919. Only long-lived radionuclides or stable nuclides are suitable as parent nuclides. To assure appropriate purity of the relevant radionuclides for medical purpose, it is necessary to establish a suitable irradiation technique as well as a proper radiochemical work-up process to assure high yield and adequate purity. This is even more important if the desired product is accessible only over more steps.

Several ways of producing the desired radionuclides have been developed. Neutron-rich nuclides either yield from nuclear fission as decay product or are created by neutron irradiation of the parent nuclides. Neutron-poor nuclides are generated using either a cyclotron or a generator.

1.2.4 Production of radioactive nuclides

1.2.4.1 Radionuclides produced by nuclear reactor^{29, 38}

Nuclear fission is a process of splitting unstable and highly excited nuclides of high atomic masses in two or more fragments of medium atomic masses and the release of energy as electromagnetic radiation and kinetic energy of the fragments. Free neutrons and protons evolve as by-product. This process of transmutation takes place in a nuclear reactor. Fission occurs spontaneously (as radioactive decay) or can be induced by a neutron flux that hits a placed element inside the reactor. The neutrons are absorbed by the element that is gaining nuclear binding energy, thus promoting to an excited state and eventually splitting into fragments. The emerging nuclides are neutron-rich and undergo beta minus decay. The most important nuclides are ^3H ($t_{1/2} = 12,3 \text{ y}$), ^{14}C ($t_{1/2} = 5730 \text{ y}$), ^{33}P ($t_{1/2} = 25,3 \text{ d}$), ^{35}S ($t_{1/2} = 87,5 \text{ d}$) and ^{125}I ($t_{1/2} = 59,4 \text{ d}$), all of which are used for scientific and medical purposes; ^3H and ^{14}C are used in liquid scintillation counting and frequently used for biochemical tasks; ^{125}I is often applied in radio immuno assays (RIA), whereas ^{33}P and ^{35}S are used in metabolic assays.

1.2.4.2 Radionuclides produced by radionuclide generator^{38, 42}

Radionuclide generators provide an economic and space-saving method of obtaining short-lived radionuclides, while solving the problem of not being able to transport short-lived radionuclides from a distant nuclear reactor to the PET facility. A parent nuclide with long half-life is used as source to obtain the daughter nuclide that can be eluted.

The generator system consists of a small glass tube filled with ion exchanger matrix retaining the parent nuclide. Both ends of the tube contain a membrane disk filtering particles. The nuclide can be eluted by attaching an evacuated injection vial.

After the daughter nuclide has been eluted, the parent begins to regenerate, so that the daughter nuclide can be obtained repeatedly.

The following prerequisites must be fulfilled to build a viable generator system:

- Half-life of the parent nuclide should be much longer than half-life of the daughter nuclide, as the lifetime of the generator is determined by the half-life of the parent nuclide
- The chemical properties of the parent nuclide must not change after repeated elution of the daughter nuclide
- The obtained daughter nuclide must provide high radiochemical purity during the lifetime of the generator system.

Popular generator systems are $^{99}\text{Mo}/^{99\text{m}}\text{Tc}$ generator, $^{81}\text{Rb}/^{81\text{m}}\text{Kr}$ generator and $^{68}\text{Ge}/^{68}\text{Ga}$ generator. $^{99\text{m}}\text{Tc}$ is widely used in single photon emission computed tomography (SPECT) due to its short half time.

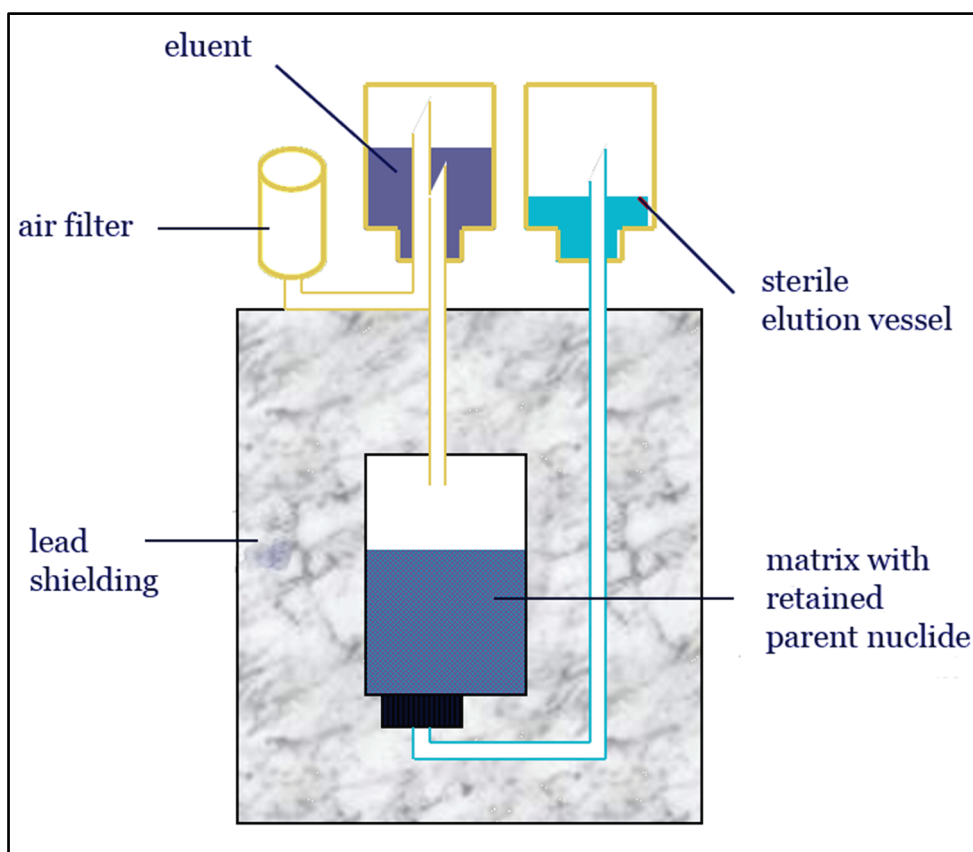


Fig.5 Generator, schematic modified⁴²

1.2.4.3 Radionuclides produced by particle accelerator (cyclotron)^{39, 42}

A cyclotron is a particle accelerator used to obtain neutron-poor radionuclides for PET imaging. In a vacuum chamber, $^1\text{H}^+$ or $^2\text{H}^+$ ions are accelerated using a high frequent alternating voltage applied. A perpendicular magnetic field forces the particles in a spiral motion. At the perimeter of the vacuum chamber, the electrons are stripped off by a stripper foil to hit the target material, which can be liquid, gaseous, or solid, causing the transmutation into the desired radionuclide. Commonly used PET nuclides are ^{18}F , ^{11}C , ^{13}N , and ^{15}O .

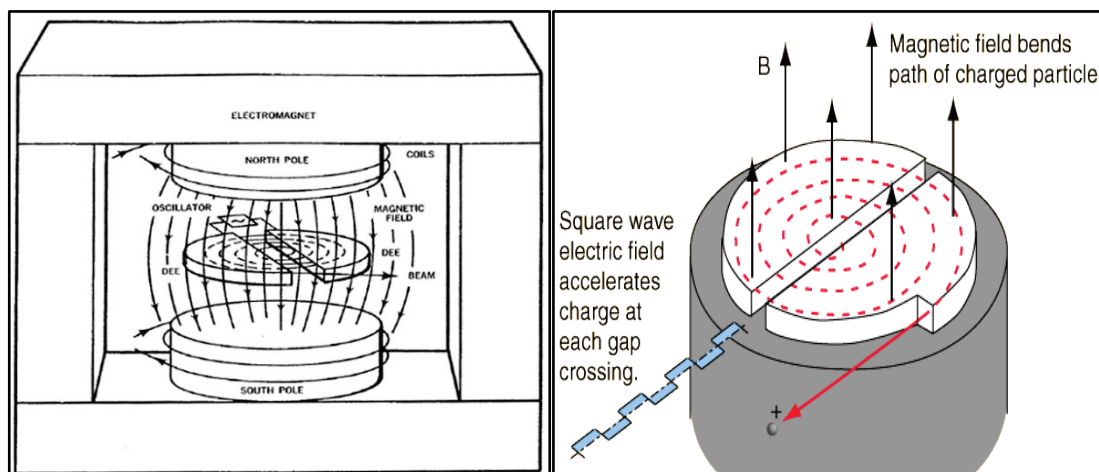


Fig.6/7 Cyclotron, schematic^{40, 41}

A cyclotron consists of a large electromagnet around a flat vacuum chamber between the two poles. Inside the chamber, a high frequency alternating voltage is applied across the „D“ electrodes (also called dees). The alternating voltage present in the gap accelerates the particles, which are injected from an ion source at the centre of the dees. The metal of the „D“ electrodes acts as Faraday shield, hence there is no electric field present inside the dees. The magnetic field runs perpendicularly to the D electrode and forces the accelerated particles to move in a spiral path. The particle beam leaves the spiral at the perimeter of the electrodes and hits the target.

1.3 Radiopharmaceuticals in functional diagnostic⁴³

Nuclear medicine exists for over 50 years and involves both therapy and diagnostics using open radionuclides. Two assumptions must be made in order to successfully employ radionuclides. First, the organism would not distinguish between the radioactive isotope from the natural occurring element; therefore it would metabolize them equally; second, the amounts of radionuclides used would be small enough that no interference with the metabolism would be observed. Thus, functional diagnostic employing radionuclides is based on a different principle than conventional medical imaging. The latter only captures static images of an organ like bones or pathological tissue, whereas functional diagnostic is able to trace a radiolabelled compound in the body over time, making its metabolism and distribution visible. Conventional nuclear imaging techniques use isotopes with short half time such as ^{123}I , ^{111}In or $^{99\text{m}}\text{Tc}$, detected by gamma camera (also called scintillation camera or Anger camera).

Recently, PET is gaining more relevance because of its superior spatial resolution. The most commonly used radio isotopes in PET are ^{18}F , ^{11}C , ^{13}N , and ^{15}O .

1.4 Positron emission tomography (PET)^{38, 44, 45, 46, 47}

Positron emission tomography (PET) is a non-invasive imaging technique, used for quantitative measurement of biological processes such as enzyme activity, receptor density, and occupancy or rate of biosynthesis in vivo. PET can be a valuable instrument in drug development and evaluation of drug efficacy. PET supported study of the adenosine receptor system is an important technique to track down neurological and behavioural diseases in the future and lead to a deeper understanding of pathways of neurotransmission. This is achieved by administration of positron emitting tracers. These are biological molecules tagged with radionuclides, favourably with no structural change of the molecules themselves. The most commonly used radio isotopes are ^{18}F ($t_{1/2} = 110$ min.), ^{11}C ($t_{1/2} = 20$ min.), ^{13}N ($t_{1/2} = 10$ min.) and ^{15}O ($t_{1/2} = 2$ min.). Due to the short half-life, it is imperative to have access to a nearby radiochemical laboratory and a

cyclotron for production purposes. The amounts of injected PET tracers are small enough (nanomolar concentration) to use highly active compounds without the risk of triggering any pharmacological effect.

1.4.1 Development of PET^{38, 47}

In 1963, approximately 10 years prior to the first use of the x-ray computed tomography (CT), Kuhl and Edwards described the Single-Photon-Emission-Computed-Tomography (SPECT). Due to limited spatial resolution of the first SPECT images, this technique was inferior to CT. At the early stages of SPECT imaging, the patient had to turn around in front of the camera, deteriorating the early images. Eventually, SPECT scanners with rotating heads have been developed, moving the camera around the patient. Today, using dual or triple headed SPECT systems is standard, mounted on a circular arc. This results in greater sensitivity and reduced scanning time as well as decreased amounts of tracer needed. These improvements combined with greater mechanical stability of modern systems decreased routine scan time from 40 to 10 minutes, making them more cost effective.

For cardiac purpose, the detectors are mounted right-angled. Moving examination beds and mobile camera tripods make dual and triple headed SPECT machines suitable for whole-body scintillation imaging.

In 1962, Rankovitz and Robertson did the first PET brain scan using a device composed of 32 NaI detectors arranged in a ring.⁴⁸

In the 1970s, the world's first commercial PET scanner was developed by Ter-Pogossian, Phelps and Hoffman⁴⁹ while Ido, Reivich and Phelps established the preparation and application of 2-deoxy-2-(¹⁸F)fluoro-D-glucose, the most widely used and versatile PET tracer.⁵⁰

1.4.2 Advantages and disadvantages of PET vs. SPECT^{47, 53, 54, 55}

PET is similar to SPECT in its use of radioactive tracers, but unlike in SPECT, gamma rays are not emitted directly by the tracer and measured by a gamma camera, but indirectly as secondary event in the annihilation process. An emitted positron travels a few millimetres before it collides with an electron, causing two gamma photons to be emitted at almost 180 degrees to each other. A PET scanner detects these photons simultaneously („in coincidence“), allowing to localize their origin along a straight line, called line of response (LOR).

In SPECT technology, the image is generated using projections from many directions, usually every 3-6 degrees recorded by a rotating camera. Acquiring sharp images requires the use of collimators, components capable of filtering a gamma ray stream, allowing only the gamma photons parallel to a certain direction to pass and hit the detector. Nevertheless, filtering over 99 % of the emitted rays has a highly negative impact on the signal-to-noise ratio leading to strong degradation of image quality.

PET, in contrary, does not use collimators, giving a better gamma photon yield, faster detection rate, better image quality, and improved spatial resolution compared to SPECT.

Furthermore, PET allows to quantifying tracer distribution patterns, which is not possible in SPECT, enabling the absolute quantification of myocardial blood flow measurement or coronary flow reserve measurement.

SPECT, however is a less pricey technology, the costs are around one third of PET machines, mainly because of two reasons. First, the scanner contains less sophisticated electronic components and second, the usage of easily obtained gamma-emitting radioisotopes with longer half-lives saves the trouble of maintaining a highly expensive cyclotron and specially adapted on-site chemical synthesis apparatus to produce the required radionuclides.

Recently, SPECT is experiencing a renaissance, having overcome some of its inherent flaws by newly developed SPECT/CT hybrid machines, while preserving its cost advantage.

Today, both SPECT and PET are used in clinical examinations, both for similar indications.

1.4.3 Application of PET⁵⁴

PET is a powerful instrument in both medicine and research. As medical instrument, it is widely used in neurology, cardiology, and oncology. Neurological diseases like Alzheimer's disease, Parkinson's disease, Chorea Huntington and epilepsy can be diagnosed in early stages and progression of these diseases can be made visible. Brain damage after a stroke can be evaluated by measuring the blood flow to different parts of the brain using ^{15}O as tracer. In the future, when more suitable radioligands for different neuroreceptor subtypes have become available (e.g. the dopamine or adenosine receptor), it is conceivable to map the brain more precisely, visualizing several kinds of brain damage arising from many neurological and psychiatric diseases, virtually undetectable by conventional SPECT/CT scans.

In cardiology, PET is helpful in studying atherosclerosis and related vascular diseases. It is also used to visualize blood flow within the coronary arteries, displaying cardiac stenosis as well as metabolic activity of the myocardium.

Most important, however, is the application of PET in clinical oncology, using 2-deoxy-2- (^{18}F) fluoro-D-glucose (commonly abbreviated ^{18}F -FDG or FDG) as standard tracer. It is a glucose analogue taken up by glucose using cells. Tumour tissue has an elevated glucose uptake due to its rapid cell proliferation rate; this is the reason why it is accumulated in cancer cells as well as in organs of high glucose utilization, such as brain or liver. Yet, a PET scan is not the method of choice to search for cancer as a routine medical check-up. That is, aside from high costs involved, because of normal inflammatory processes that lead to accelerated glucose metabolism as well, hence cannot be distinguished from tumour tissue in a PET image. Nevertheless, metastasis can be made visible, making it an ideal instrument for staging and monitoring of cancer. It is also used to assess cancer treatment success, particularly of Hodgkin's lymphoma, non-Hodgkin's lymphoma, lung cancer and several other kinds of solid tumours.

The role of PET in pharmacology includes biodistribution studies in pre-clinical trials, when radiolabelled new drugs are injected into small animals to trace a possible new drug inside the body. Distribution, tissue concentration, and elimination of the compound can be discovered very quickly and cost-effective.

Moreover, drug occupancy at a specified target can be inferred indirectly by competition studies between unlabelled drug and a radiolabelled compound known to bind.

1.4.4 Hybrid Technology⁵⁴

Recently, large efforts were made to combine the best of both worlds, by coupling SPECT and/or PET with conventional CT. The latter is used to calculate the attenuation map, necessary to correct for the photons absorbed by overlying tissue. Additionally, the CT provides valuable anatomical information, while PET gives the physiological information. The result is a more detailed image by merging the two, enabling a more accurate diagnosis, as well as shortening scan time. The increased accuracy and image quality of PET/CT or SPECT/CT machines can also help avoiding unnecessary surgery; it allows better determination of patient's response to therapy and more precise guiding of radiation therapy. Newly developed PET/CT hybrids are also capable of taking 4D images, which means taking images of moving anatomical structures, such as heart or lungs.

1.4.5 Production of [^{18}F]-radiolabelled synthesis precursors for PET^{56, 57, 58}

The successful development of a novel PET tracer requires observing following aspects:

- choice of labelling position in the tracer molecule
- stereochemical properties
- specific activity
- choice of the cyclotron produced primary precursor and online production of the secondary precursor
- chemical properties of the precursor, including the choice of the solvent
- choice of solvent for the pharmaceutical formulation (pH adjustment to physiological state and osmolality)
- expected clinical outcome of the nuclear medicinal question

One important aspect of developing novel PET tracers is the specific activity of the radioligand. The specific activity is defined as ratio of the radiolabelled molecules to the total number of molecules. The higher the specific activity of a radioligand is, the fewer molecules need to be applied for the PET imaging process and the less interference with the observed biological process occurs. This is of great significance in measuring receptor density and distribution patterns as well as enzyme level tests.⁵⁷

Production of all PET radiotracers starts with small cyclotron obtained precursor molecules. Yet, the number of the resulting precursor molecules of the four commonly used radionuclides ^{11}C , ^{18}F , ^{13}N , and ^{15}O is limited due to extreme conditions in the cyclotron target. As the chemical form is determined by high energy, the irradiation leads to stable and rather unreactive molecules. There are many ways of producing the desired radionuclides, but usually only one or two different nuclear reactions are favoured.

The ^{18}F nucleus is the most relevant in this thesis and usually produced in either of two ways.⁵⁹ One is the $^{20}\text{Ne}(\text{d}, \alpha)^{18}\text{F}$ nuclear reaction on a target of pressurized neon gas using a nickel target body to obtain $[^{18}\text{F}]\text{F}_2$, the diatomic and highly reactive species of ^{18}F .⁶⁰

The other, more widely used is the $^{18}\text{O}(\text{p}, \text{n})^{18}\text{F}$ nuclear reaction on a H_2^{18}O target using a target body of pure silver or titan. After irradiation, ^{18}F is obtained as fluoride ion ($[^{18}\text{F}]\text{F}^-$) in aqueous solution and can be used for further reactions in aqueous solution or can be separated from H_2^{18}O by distillation or using ion exchanger resin.^{61, 62, 63}

1.5 ^{18}F radiolabelling techniques

1.5.1 Nucleophilic fluorination⁶⁴

The $^{18}\text{F}^-$ ion derived from the aqueous solution is used for nucleophilic fluorination. Nevertheless, the fluoride ion is unreactive, because of its high charge density and the large surrounding hydration shell. In order to remove H_2^{18}O and interfering metal ions from the target body, anionic exchanger resin is used.^{65, 66, 67} Then the $^{18}\text{F}^-$ is eluted by a solution of K_2CO_3 and the cryptand „Kryptofix 2.2.2TM“ in an acetonitrile/water mixture (7+3). After the elution, additional acetonitrile is added to get an acetonitrile/water mixture (85+15), which can be removed easily by azeotropic distillation under an inert gas stream. The cryptand complexing the $^{18}\text{F}^-$ can then be transferred to a suitable organic solvent for further reaction.

The displacement of different leaving groups by $^{18}\text{F}^-$ is an excellent choice of forming aliphatic Carbon-Fluor bonds.⁶⁸ Selecting a suitable leaving group depends on accessibility of the molecule, stability of the precursor, the method of isolating the ^{18}F labelled product, reagents, solvent, and formation of by-products. Especially trifluoromethanesulfonate esters are characterized by high reactivity and lead to high yields in nucleophilic fluorination as in the preparation of 2-deoxy-2- ^{18}F fluoro-D-glucose. Halides such as Cl^- , Br^- , I^- are also good leaving groups and widely used, whereas tosylate and mesylate groups sometimes are a bit lower yielding.⁵⁶

1.5.2 Electrophilic fluorination⁶⁴

The simplest way of fluorination via electrophilic attack by ^{18}F is the preparation of $[^{18}\text{F}]\text{F}_2$ as described above. The most significant problem of using the diatomic species, however, is its extremely high reactivity, associated with low selectivity. The exothermic reactions need appropriate cooling or the use of highly diluted ^{18}F solutions. A more feasible way is the conversion into less reactive functional groups such as in acetyl hypofluorite⁶⁹ or xenon difluoride⁷⁰. Despite its disadvantages, the electrophilic fluorination is well established in PET tracer development.

1.5.3 Fluoroalkylations^{56, 71}

Many biologically active compounds contain alkyl side chains like methyl or ethyl groups suitable for radionuclide labelling. Since there are many $[^{14}\text{C}]$ methyl tracers available, development of $[^{18}\text{F}]$ alkylated analogues was the next logical step. To do so, a number of $[^{18}\text{F}]$ alkylating agents were developed. $[^{18}\text{F}]$ bromo fluoromethane, $[^{18}\text{F}]$ fluoro iodomethane, 2- $[^{18}\text{F}]$ bromo fluoroethane, 2- $[^{18}\text{F}]$ tosyloxy fluoroethane are a few to mention. The radioligands $[^{18}\text{F}]$ FET (O-(2- $[^{18}\text{F}]$ fluoroethyl)L-tyrosine) and $[^{18}\text{F}]$ FP-CIT (*N*-(3- $[^{18}\text{F}]$ fluoropropyl)-2 β -carbomethoxy-3 β -(4-iodophenyl)nortropine) are already in clinical use.

1.5.3.1 Fluoroethylations

Fluoroethylations are the most relevant among the fluoroalkylations, because fluoroalkylation reagents can be produced easily by low priced and widely available precursors. Additionally, the fluoroethyl group strongly resembles both the methyl and ethyl group. Popular targets for fluoroethylations are amine-, hydroxyl-, mercapto- and carboxyl groups, using reagents such as $[^{18}\text{F}]$ tosyloxy fluoroethane, $[^{18}\text{F}]$ fluoro ethyl trifluoromethanesulfonate or $[^{18}\text{F}]$ tosyloxy fluoroethane.

2. MY OWN RESEARCH

2.1 Aim of my research

Based on the results of Li et al⁷⁴, who investigated the selective binding of pyridine derivatives as antagonists to the adenosine A₃ receptors, [¹⁸F]FE@SUPPY seems to be an appropriate candidate to act as selective radiotracer of high affinity.⁷⁴ This compound has a **sulfanyl-phenyl-pyridine** structure of which derives the abbreviation SUPPY and is radiolabelled using ¹⁸F on the carboxyl group. A structural related compound is [¹⁸F]FE@SUPPY:2, which is radiolabelled on the thiocarboxylic side, expected to yield an even better receptor affinity. Many related compounds have been synthesized to date, trying to modify the lipophilicity of the SUPPY molecule in order to achieve better receptor binding quality.

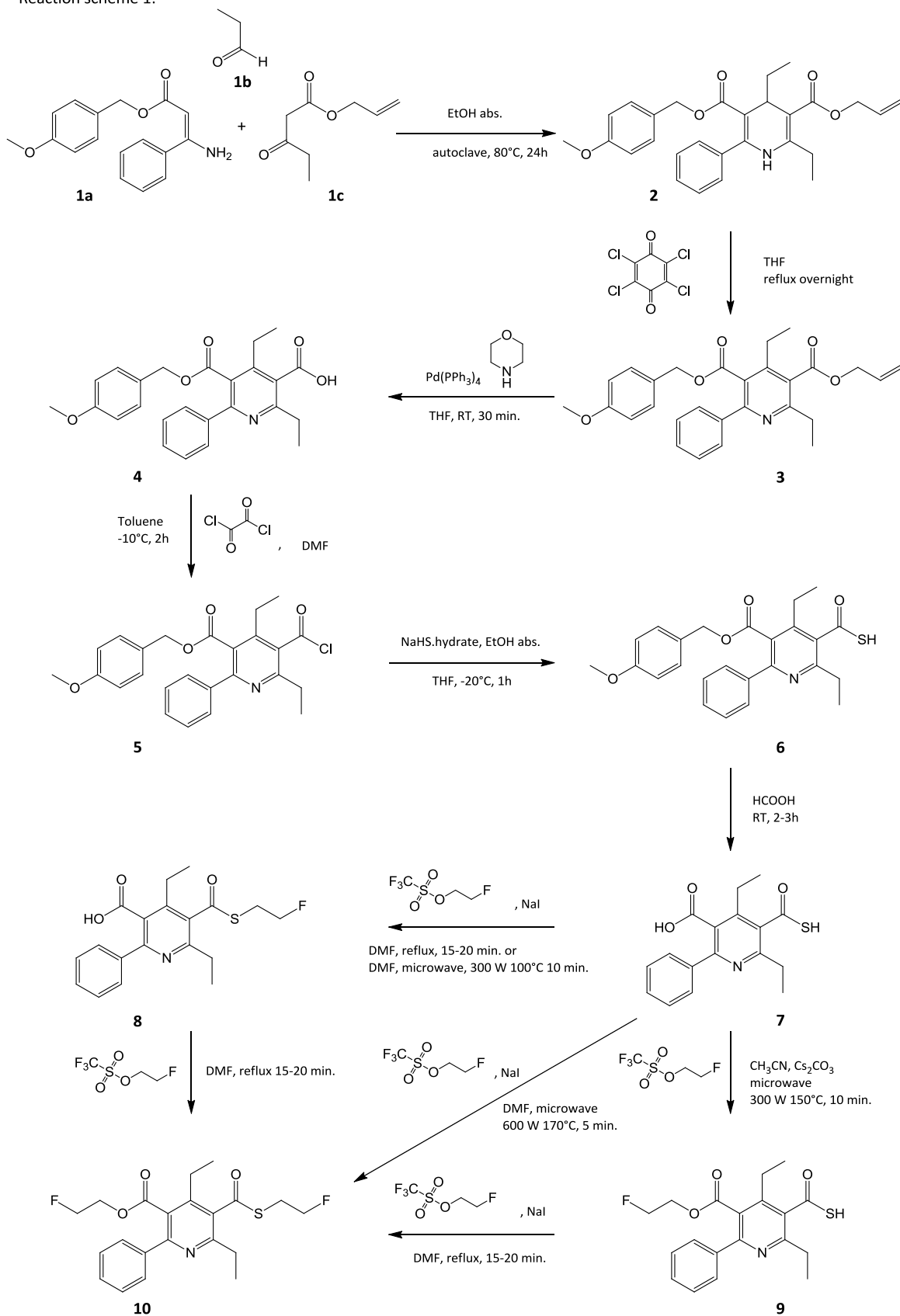
The aim of my research was the preparation of two novel SUPPY derivatives. They are 2-fluoroethyl 4,6-diethyl-5-[[(2-fluoroethyl)thio]carbonyl]-2-phenylnicotinate (**10**) and methyl 4,6-diethyl-5-[[(2-fluoroethyl)thio]carbonyl]-2-phenylnicotinate (**11**).

Both compounds should serve as HPLC reference compounds and are candidates to be tested as novel PET tracers for the adenosine-A₃-receptor.

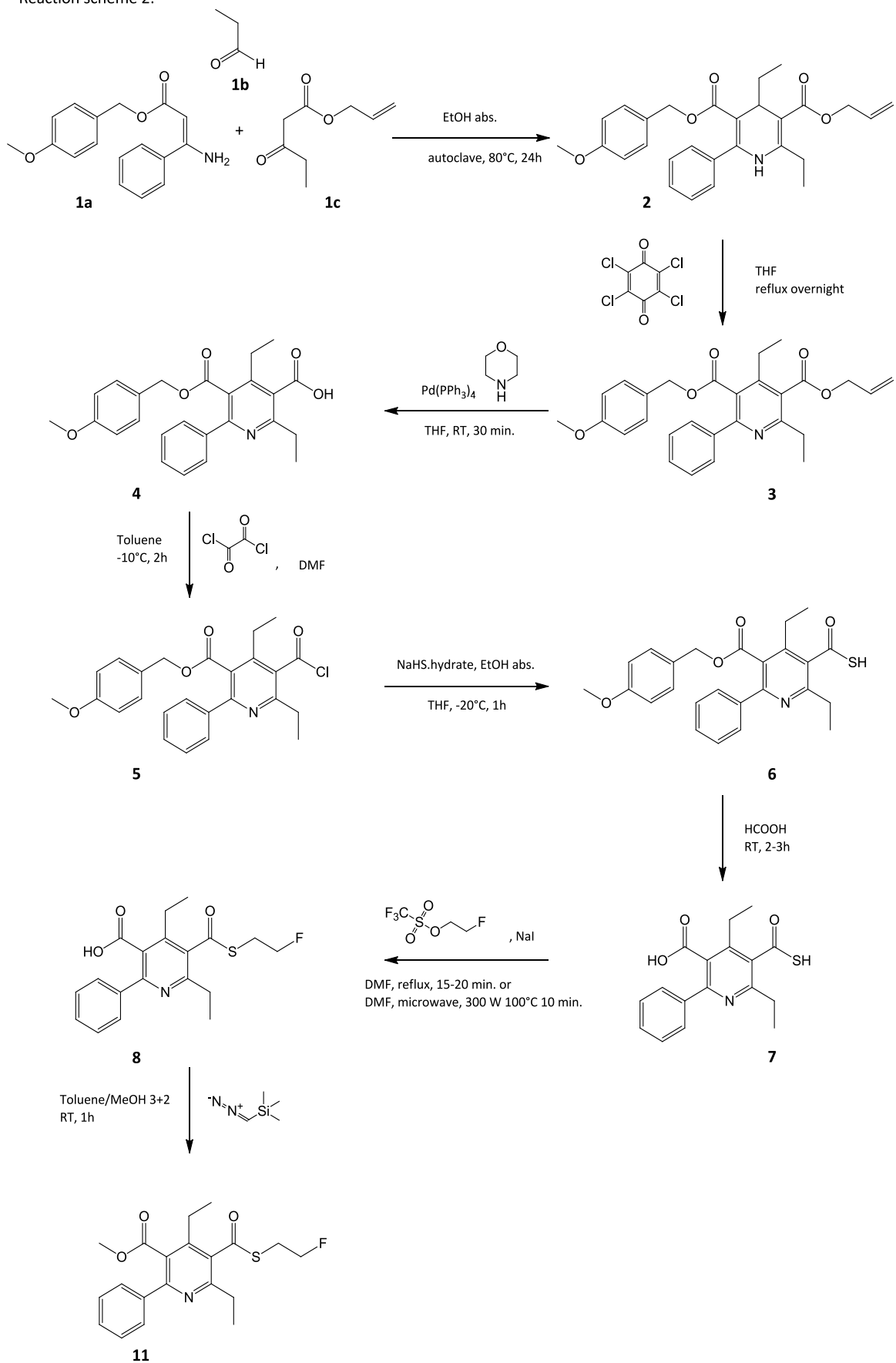
Furthermore, a new synthesis pathway to obtain 4,6-diethyl-5-[[(2-fluoroethyl)thio]carbonyl]-2-phenylnicotinic acid (**8**) was established.

2.2 Reaction scheme

Reaction scheme 1:



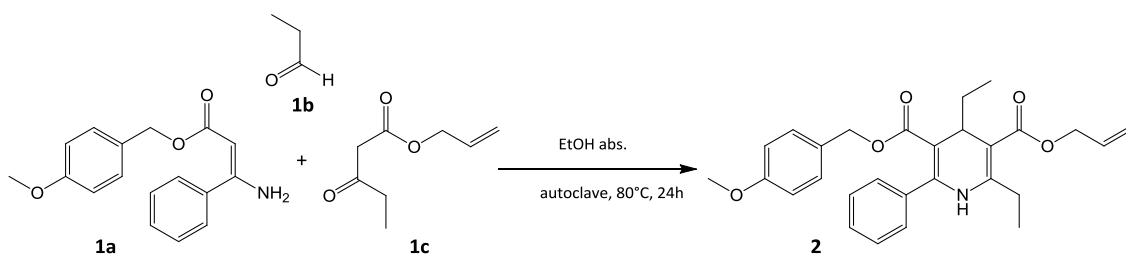
Reaction scheme 2:



2.3 Preparation of 4,6-diethyl-2-phenyl-5-thiocarboxynicotinic acid⁷¹ (**7**)

The first desired compound was 4,6-diethyl-2-phenyl-5-thiocarboxynicotinic acid (also called SUPPY:o) (**7**), which can be considered the parent compound, allowing quick and easily performed modification with different side chains. SUPPY:o alterations of the basic structure led to several PET precursors, of which some are already in use. The first six steps were carried out according to the dissertation of Shanab K., 2007, with some procedure modifications in order to increase the yield of SUPPY:o.

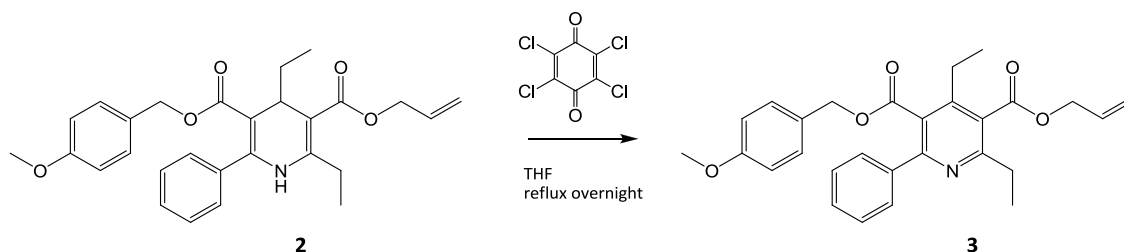
2.3.1 Preparation of 3-allyl 5-(4-methoxybenzyl) 2,4-diethyl-6-phenyl-1,4-dihydropyridine-3,5-dicarboxylate⁷¹ (**2**)



In the first step, p-methoxybenzyl-protected and allyl-protected precursors were subjected to a Hantzsch pyridine synthesis. 4-Methoxybenzyl-3-amino-3-phenylprop-2-enoate (**1a**), allyl-3-oxopentanoate (**1c**), and propionaldehyde (**1b**) were dissolved in absolute ethanol and stirred in the autoclave at 80°C for 24 hours. The solvent was removed after cooling to room temperature and the crude product purified by liquid column chromatography eluting with ligroine/ethyl acetate 8+2.

2.3.2 Preparation of 3-allyl 5-(4-methoxybenzyl) 2,4-diethyl-6-phenylpyridine-3,5-dicarboxylate⁷¹ (**3**)

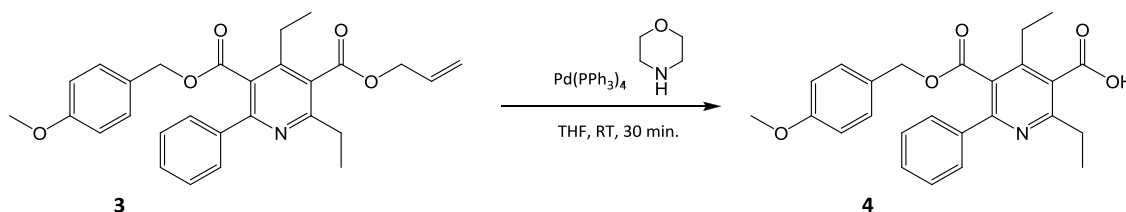
The second step employs the oxidization of (**2**) to the desired pyridine derivative (**3**), according to following scheme.



3-Allyl 5-(4-methoxybenzyl) 2,4-diethyl-6-phenyl-1,4-dihydropyridine-3,5-dicarboxylate (**2**) and 2,3,5,6-tetrachloro-1,4-benzoquinone were dissolved in THF and heated under reflux overnight. The reaction mixture was cooled down and the solvent removed under reduced pressure to give the desired pyridine derivative (**3**). The product was purified by using column chromatography eluting with ligroine/ethyl acetate 9+1.

2.3.3 Preparation of 2,4-diethyl-5-[(4-methoxybenzyl)oxy] carbonyl}-6-phenylnicotinic acid⁷¹ (**4**)

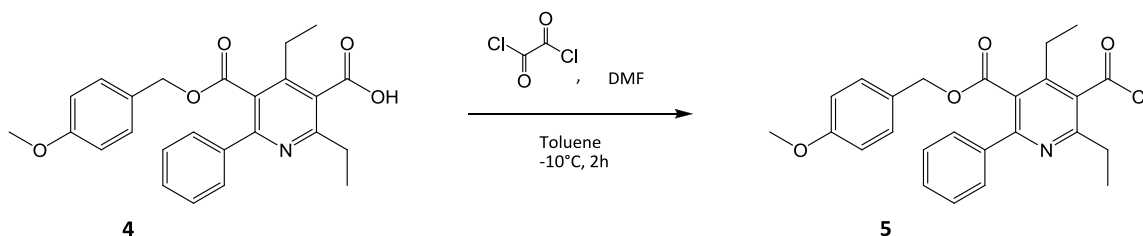
In the following step, the allyl protecting group was removed. The allyl ester function could be split off selectively by the use of transition metal tetrakis(triphenylphosphine)palladium and morpholine.



3-Allyl 5-(4-methoxybenzyl 2,4-diethyl-6-phenylpyridine-3,5-dicarboxylate (**3**), obtained from the previous step was dissolved in THF and then tetrakis(triphenylphosphine)palladium and morpholine were added under argon atmosphere. The mixture was stirred for 30 minutes at room temperature. Afterwards, the solvent was removed under reduced pressure. The obtained product required extensive purification operations. It was purified by liquid chromatography (silica gel 60) several times. First, an eluent mixture of dichloromethane/methanol 95+5 was used for separating the dark brown contamination from the product. Afterwards, an ethyl acetate/methanol 8+2 mixture was used to elute the product faster. Several repetitions were necessary to achieve a satisfactory result.

2.3.4 Preparation of 4-methoxybenzyl 5-(chlorocarbonyl)-4,6-diethyl-2-phenylnicotinate⁷¹ (**5**)

The product from the previous step (**4**) was used for converting the carboxylic acid group into the corresponding acid chloride (**5**).



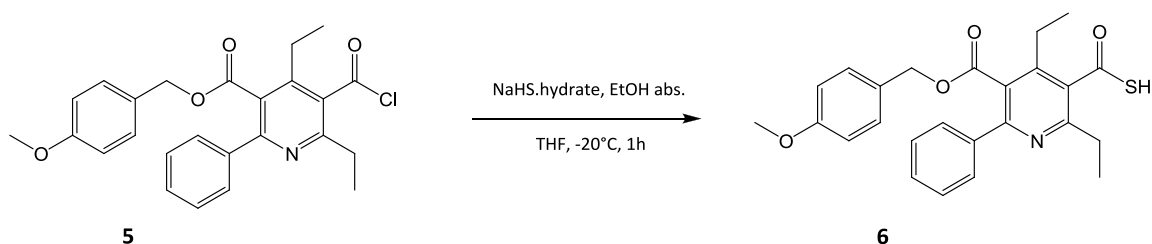
To perform this, 2,4-diethyl-5-[(4-methoxybenzyl)oxycarbonyl]-6-phenylnicotinic acid (**4**) was dissolved in absolute toluene and injected via syringe to a three-necked-flask equipped with reflux condenser and thermometer under inert gas. The apparatus was then immersed in a Dewar vessel filled with a ethanol/dry ice mixture and cooled down to -20°C instead of -10°C of the original working instructions, hoping of an yield increase of the desired thiocarboxylate. Another modification was the usage of toluene instead of benzene as solvent mainly for two reasons. First, toluene is less toxic and second, toluene's freezing temperature is below that of benzene, assuming the reaction mixture would stay

liquid during the whole reaction. The more difficult removal of toluene at room temperature proved to be the only disadvantage over benzene.

Then a few drops of DMF were added as catalyst and the oxalyl chloride injected drop wise via syringe. After removing the Dewar and reaching room temperature, the mixture was stirred for additional 30 minutes, eventually becoming a brownish liquid and stirred until the reaction was finished after approximately one hour. Afterwards, the solvent was removed under reduced pressure at room temperature.

2.3.5 Preparation of 2,4-diethyl-5-[(4-methoxybenzyl)oxy] carbonyl}-6-phenylpyridine-3-carbothioic S-acid⁷¹ (6)

The product obtained from the previous step (5) was subjected to the following reaction to give the desired thiocarboxylic acid.



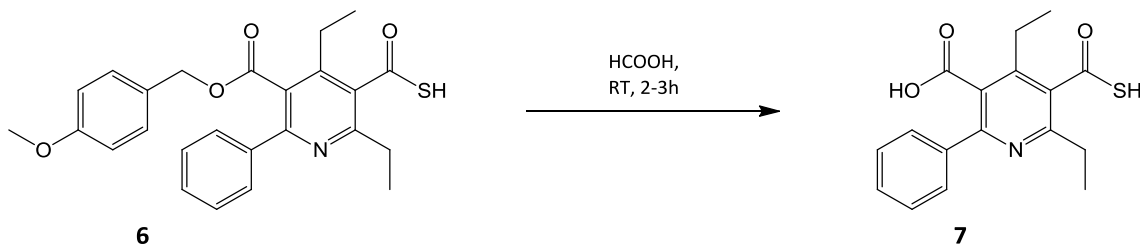
In a dry three-necked flask equipped with reflux condenser under inert gas atmosphere, comminuted NaHS·hydrate platelets were dissolved in absolute ethanol under moderate heating. After having become completely dissolved, the solution was immersed in a Dewar vessel filled with a ethanol/dry ice mixture and cooled down to -20°C, hoping the additional cooling, as in the step before, would help to increase the SUPPY:o yield. Unfortunately, the attempt proved to be unsuccessful in both cases.

The 4-methoxybenzyl 5-(chlorocarbonyl)-4,6-diethyl-2-phenylpyridine-3-carboxylate (5) residue yielding from the previous reaction was dissolved in absolute THF and then added slowly via syringe under stirring. After the addition of the acid chloride had finished, the Dewar vessel was removed, allowing the reaction mixture to reach room temperature. The stirring was continued for a further hour,

during which the progress of the reaction was monitored by TLC. Afterwards, the orange solution was acidified by 6N hydrochloric acid. The aqueous layer was extracted thoroughly with ethyl acetate, washed twice with a saturated solution of sodium chloride, and dried over sodium sulphate. The solvent was removed under reduced pressure and the crude product purified by column chromatography several times eluting with a ligroine/ethyl acetate mixture 7+3. Almost colourless crystals were obtained.

2.3.6 Preparation of 4,6-diethyl-2-phenyl-5-thiocarboxynicotinic acid⁷¹(**7**)

The final step in the preparation of SUPPY:o (**7**), the parent compound for novel PET tracer preparation, comprises of cleaving the p-methoxybenzyl protection group.

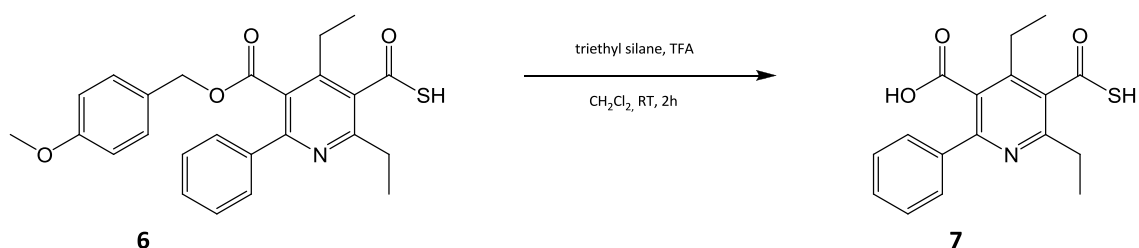


For the desired deprotection, 2,4-diethyl-5-([(4-methoxybenzyl)oxy]carbonyl}-6-phenylpyridine-3-carbothioic S-acid (**6**) from the previous step was stirred in the presence of formic acid at room temperature for several hours. Then the formic acid was removed under reduced pressure. The crude product was purified by reversed-phase column chromatography several times, eluting with an acetonitrile/water mixture 95+5. Compound (**7**) was obtained as colourless crystals.

Unfortunately, this purification was hampered by a considerable reduction of yield (41%). Two alternate ways of cleaving the protection group were investigated in order to prevent this substantial loss of product.

2.3.6.1 Deprotection with triethyl silane / trifluoroacetic acid⁷³

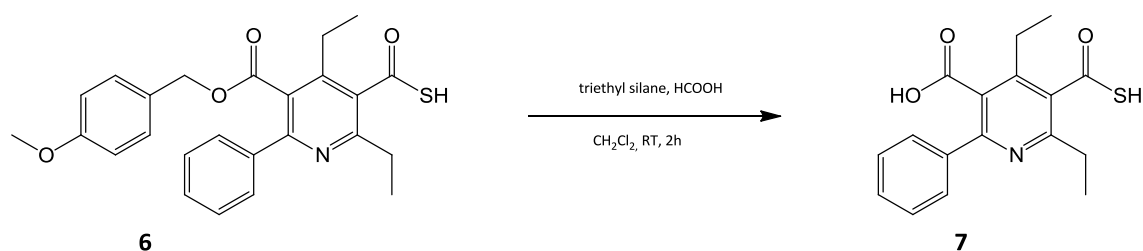
In order to prevent the substantial loss of SUPPY:O during the deprotecting step, a different approach was tried using triethyl silane serving as carbocation scavenger.



2,4-Diethyl-5-([(4-methoxybenzyl)oxy]carbonyl)-6-phenylpyridine-3-carbothioic S-acid (**6**) was dissolved in dry dichloromethane in a three-necked-flask equipped with reflux condenser and thermometer under argon atmosphere and cooled down to 0°C in an ice bath. Then triethyl silane and trifluoroacetic acid were added via syringe. A considerable rise in temperature was not noticed. The ice bath was removed and the solution stirred for further two hours. The colour of the solution turned dark reddish-brown. The solvent was evaporated under reduced pressure and the crude product was purified by reversed-phase column chromatography several times, eluting with an acetonitrile/water mixture 95+5. Only about 41% of highly purified SUPPY:O (**7**) resulted from this attempt. Considering the effort necessary to perform this preparation, this method is not preferable over the originally published method.

2.3.6.2 Deprotection with triethyl silane / formic acid⁷³

Based on the observation that in the previous attempt the reaction mixture turned darker during the reaction, it was suspected that TFA was a too strong deprotecting agent and would possibly lead to decomposition of the product. In consequence of this, TFA was replaced by formic acid, expecting an increase of yields of SUPPY:o (**7**).



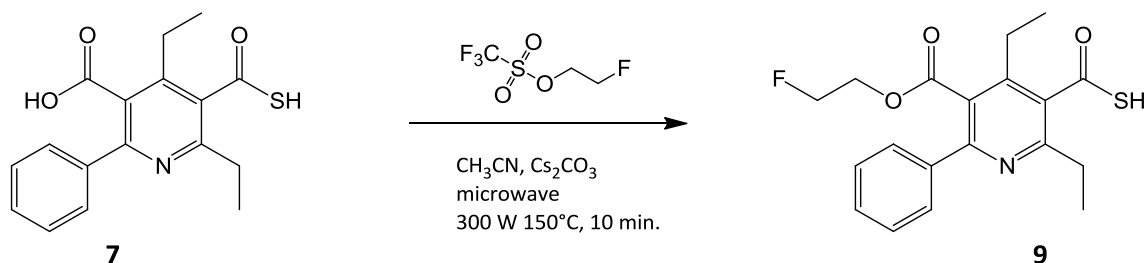
2,4-Diethyl-5-[[[4-methoxybenzyl]oxy]carbonyl]-6-phenylpyridine-3-carbothioic S-acid (**6**) was dissolved in dry dichloromethane in a three-necked-flask equipped with reflux condenser and thermometer under argon atmosphere and cooled to 0°C in an ice bath. Then triethyl silane and formic acid were added via syringe. No considerable rise in temperature was observed. The ice bath was removed and the solution stirred for further two hours. The colour of the solution changed to yellow. Afterwards, the solvent was evaporated under reduced pressure and the crude product was purified by reversed-phase column chromatography eluting with an acetonitrile/water mixture 95+5. Only about 19% of SUPPY:o (**7**) was obtained from this attempt.

2.4 Preparation of 2-fluoroethyl 4,6-diethyl-5-[(2-fluoroethyl)thio]carbonyl}-2-phenylnicotinate (10)

The preparation of (9) starting from SUPPY:o (7) can be achieved by 3 different routes:

1. 2-fluoroethylation of the carboxylate function prior to the 2-fluoro ethylation of the thiocarboxylate function
2. 2-fluoroethylation of the thiocarboxylate function prior to the 2-fluoro ethylation of the carboxylate function
3. In a microwave-assisted single-step reaction

2.4.1 Microwave-assisted preparation of 2,4-diethyl-5-[(2-fluoroethoxy)carbonyl]-6-phenylpyridine-3-carbothioic S-acid⁷¹ (9)



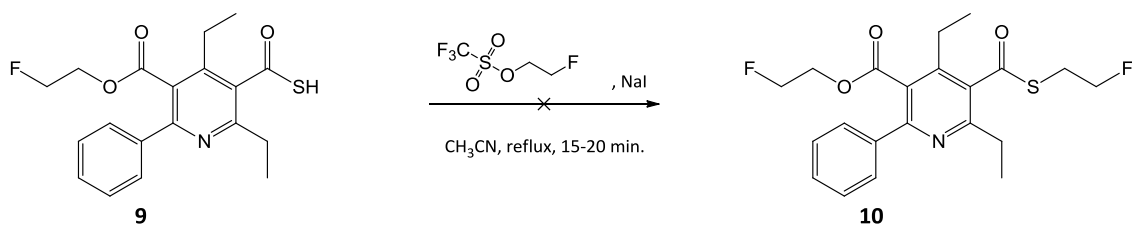
4,6-Diethyl-2-phenyl-5-thiocarboxynicotinic acid (**7**), 2-fluoroethyl trifluoromethanesulfonate and caesium carbonate were suspended in acetonitrile in a quartz glass vessel and put to the reaction chamber. The microwave parameters were set to 300 W, 150°C and 10 minutes reaction time. Caesium carbonate serves for two purposes, as a dipole supposed to superheat the solvent and acting as base to deprotonize the carboxylic acid.

Caused by a defective temperature sensor the reaction was abrupted after 2:36 minutes. Nevertheless, about 70 % product was obtained. In prior attempts⁷¹, a

total reaction time of 10 minutes was sufficient to get about 90 % yield. The solvent was removed under reduced pressure and the crude product purified by reversed-phase column chromatography eluting with acetonitrile.

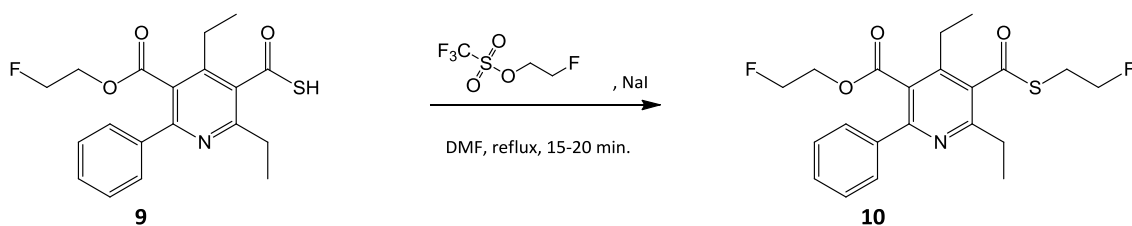
2.4.2 Preparation of 2-fluoroethyl 4,6-diethyl-5-[(2-fluoroethyl)thio]carbonyl}-2-phenylnicotinate (10)

The second step comprises the fluoroethylation of the product obtained in the previous step.



2,4-Diethyl-5-[(2-fluoroethoxy)carbonyl]-6-phenylpyridine-3-carboxylic acid (**9**) obtained from the previous step, sodium iodide and 2-fluoroethyl trifluoromethanesulfonate were suspended in acetonitrile in a round-bottomed flask equipped with reflux condenser and heated under reflux for 15-20 minutes. Acetonitrile, however, proved to be an unsuitable solvent, because no reaction progress could be observed. Furthermore, the addition of caesium carbonate as in the step before did not lead to a successful reaction.

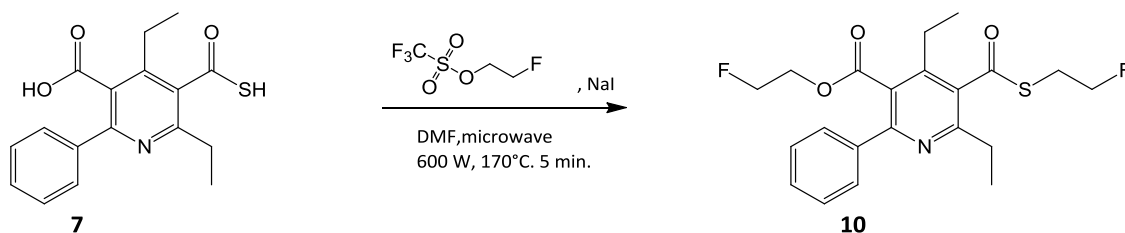
Although the use of DMF as solvent is accompanied by some disadvantages, it should replace the inappropriate acetonitrile.



As a consequence, 2,4-diethyl-5-[(2-fluoroethoxy)carbonyl]-6-phenylpyridine-3-carbothioic S-acid (**9**) from the step earlier, sodium iodide and 2-fluoroethyl trifluoromethanesulfonate were dissolved in DMF in a round-bottomed flask equipped with reflux condenser and heated for 15-20 minutes under reflux. No adding of caesium carbonate was necessary. The reaction mixture had a dark orange colour. Then the solvent was removed by ball tube distillation and additional drying on a vacuum pump. The crude product was purified several times by reversed-phase column chromatography eluting with an acetonitrile/water mixture 9+1. Highly viscous orange oil was obtained. NMR and high-resolution mass spectra revealed the successful preparation of the product.

2.4.3 Microwave assisted single-step preparation of 2-fluoroethyl 4,6-diethyl-5-[(2-fluoroethyl)thio]carbonyl}-2-phenylnicotinate (**10**)

During the attempt of the microwave-assisted preparation of 4,6-diethyl-5-[(2-fluoroethyl)thio]carbonyl}-2-phenylnicotinic acid (**8**), a lucky coincidence occurred to give the product 2-fluoroethyl 4,6-diethyl-5-[(2-fluoroethyl)thio]carbonyl}-2-phenylnicotinate (**10**) in one single step.



4,6-Diethyl-2-phenyl-5-thiocarboxynicotinic acid (**7**), sodium iodide and 2-fluoroethyl trifluoromethanesulfonate were dissolved in DMF in a quartz glass vessel. Microwave power was set to 600 W, thus doubling the usual power setting and the temperature limit was set to 170°C and 5 minutes reaction time. These parameters obviously resulted in preferring of the product (**10**) over (**8**). Most remarkably, neither additional amount of 2-fluoroethyl trifluoromethanesulfonate

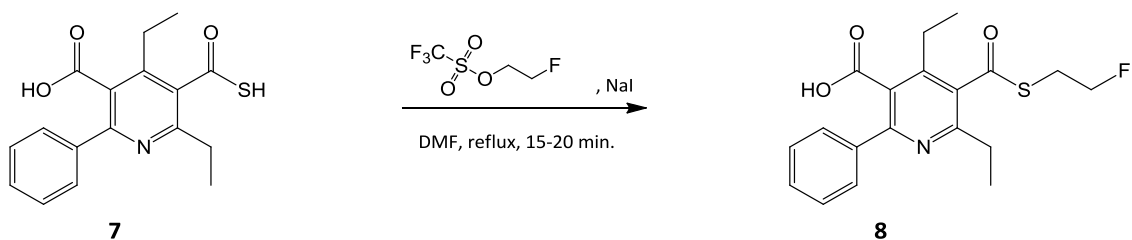
nor adding of caesium carbonate was necessary to lead successfully to product **(10)**.

The reaction mixture was of light yellow colour. The solvent was removed by ball tube distillation and additional drying on a vacuum pump. The crude product was purified several times by reversed-phase column chromatography eluting with acetonitrile/water 9+1. Compound **(10)** was obtained as highly viscous oil.

2.5 Preparation of methyl 4,6-diethyl-5-{[(2-fluoroethyl)thio]carbonyl}-2-phenylnicotinate (**11**)

The preparation of desired compound **(11)** was achieved in two steps starting from SUPPY:o (**7**). First the thiocarboxylate group was fluoroethylated followed by the methylation of the carboxylate function. Furthermore, the first step was performed using conventional preparation and microwave assisted preparation in order to compare yield rates.

2.5.1 Preparation of 4,6-diethyl-5-{[(2-fluoroethyl)thio]carbonyl}-2-phenylnicotinic acid (**8**)

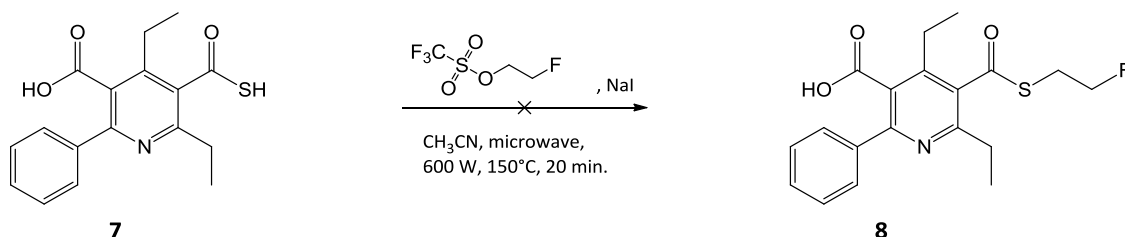


4,6-Diethyl-2-phenyl-5-thiocarboxynicotinic acid (**7**), sodium iodide and 2-fluoroethyl trifluoromethanesulfonate were dissolved in DMF in a round-bottomed flask equipped with reflux condenser and heated for 15-20 minutes under reflux. During this time, the reaction mixture turned to orange colour. The solvent was removed by ball tube distillation. The residue was dissolved in ethyl acetate and alkalized with a 2N sodium hydroxide solution. The organic layer was extracted with water and the aqueous layer was concentrated under reduced

acidified with 2N hydrochloric acid, and thoroughly extracted with ethyl acetate. The solvent was removed under reduced pressure and the crude product purified by reversed-phase column chromatography eluting with acetonitrile/water 9+1. Colourless crystals of **(8)** were obtained. Pre-purification by extraction was not necessary employing the conventional method; nevertheless, it is a reasonable method leading to a pure product.

2.5.2 Microwave-assisted preparation of 4,6-diethyl-5-[(2-fluoroethyl)thio]carbonyl}-2-phenylnicotinic acid (**8**)

As in the step of the microwave-assisted fluoroethylation of the carboxylic acid (see 2.4.1.), it was attempted to fluoroethylate the thiocarboxylate function selectively. Trying to avoid the tedious purification steps using the solvent DMF and the previous successful attempt of the preparation of **(9)** were the reason to select acetonitrile as solvent.

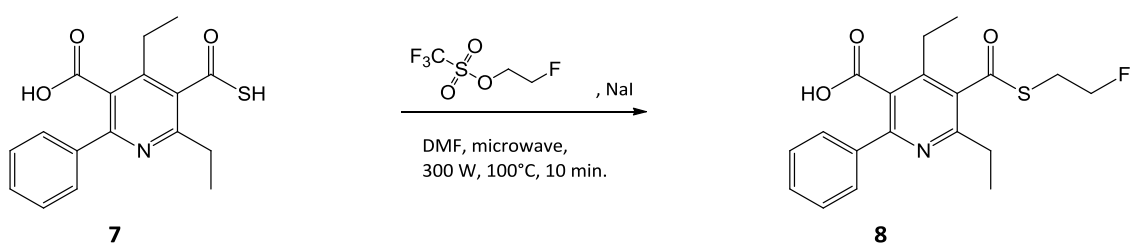


As a consequence, 4,6-diethyl-2-phenyl-5-thiocarboxynicotinic acid **(7)**, 2-fluoroethyl trifluoromethanesulfonate and sodium iodide were suspended in acetonitrile in a quartz glass vessel. Microwave parameters were set to 600 W, 150°C and 10 minutes reaction time. After 10 minutes almost no conversion of the reactant had been noticed, so it was decided to continue for further 10 minutes. Unfortunately, TLC monitoring did not show any successful reaction.

In the second attempt, the solvent was replaced by DMF (all microwave parameters remained unchanged). This time the reactant had been converted almost completely, but only the “wrong” product **(10)** was obtained (see 2.4.3). This lucky coincidence caused me to change the microwave parameters.

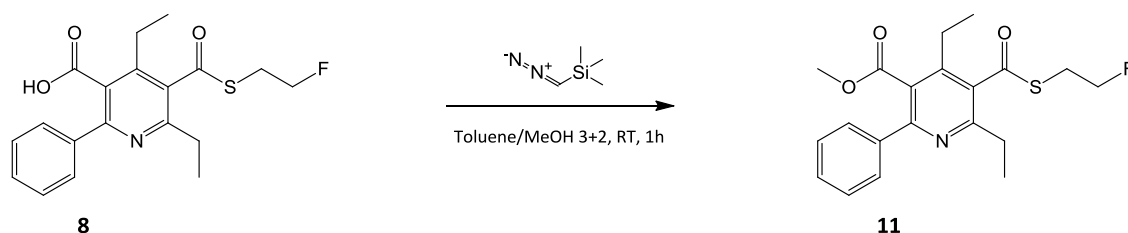
Power setting was now adjusted to 300 W and the temperature limit was decreased to 100°C. These settings proved to be successful as shown below.

After the reaction, the solvent was removed under reduced pressure. The residue was dissolved in ethyl acetate and alkalized with a 2N sodium hydroxide solution. The organic layer was extracted with water. The aqueous layer was concentrated under reduced pressure, acidified with 2N hydrochloric acid, and thoroughly extracted with ethyl acetate. The solvent was removed under reduced pressure and the crude product purified by reversed-phase column chromatography eluting with acetonitrile/water 9+1.



However, scaling up of this reaction showed some unexpected consequences. First, the yield of product was lower. Furthermore, increasing the reaction time did not show any effect. Some of the reactant remained unchanged. The more worrying disadvantage occurred during the purification process. Some contamination virtually undetectable on TLC sheet, but detectable via NMR spectra required additional purification steps. A pre-purification was carried out by extracting several times. Column chromatography using different eluent combinations (acetonitrile/water 9+1, ethyl acetate, dichloromethane/methanol 95+5) was performed. Yet, the contamination could not be entirely eliminated. Considering the time necessary for making preparations for use of the microwave oven and the following tedious purification process, the microwave-assisted approach is not favourable over the conventional route.

2.5.3 Preparation of methyl 4,6-diethyl-5-[[(2-fluoroethyl)thio] carbonyl]-2-phenylnicotinate⁷² (**11**)



4,6-Diethyl-5-[[(2-fluoroethyl)thio] carbonyl]-2-phenylnicotinic acid (**8**), obtained from either the reaction at 2.5.1. or 2.5.2 was dissolved in a mixture of toluene/methanol 3+2 in a round-bottomed flask and stirred.

(Diazomethyl)trimethylsilane solution in hexane was added under continuous stirring until the yellow colour persisted. The stirring was continued for 30 minutes. The progress of the reaction was observed via TLC monitoring. Additional (diazomethyl)trimethylsilane solution was added and the stirring continued. After an overall stirring time of one hour, no further progress could be observed. The solvent was removed under reduced pressure and the crude product was purified by reversed-phase column chromatography, eluting with acetonitrile/water 9+1.

Compound (**11**) was obtained as colourless crystals.

The contaminated reactant (**8**) of the microwave-assisted approach yielded only about 25%, a further reason why not to recommend the microwave assisted preparation of (**8**).

3. EXPERIMENTS

3.1 Technical Equipment

Recording of spectra:

Mass spectra: Shimadzu QP 1000 (EI, 70eV), direct inlet

High resolution mass spectra: Finnigan MAT 8230 (EI, 70eV) and Finnigan MAT 900 S (ESI, 4 kV, 3 μ A CH₃CN/MeOH).

¹H, ¹³C NMR-Spectra: Bruker Avance DPX-200 spectrometer at 27 °C (200.13 MHz for ¹H, 50.32 MHz for ¹³C). The solvent signal is used as internal reference standard related to TMS, occurring at δ = 7.26 ppm (¹H in CDCl₃), δ = 2.49 ppm (¹H in DMSO-*d*₆), δ = 77.0 ppm (¹³C in CDCl₃) and δ = 39.5 ppm (¹³C in DMSO-*d*₆).

Infrared spectra: Perkin-Elmer FTIR spectrometer Spectrum 1000 (KBr pellets).

Chromatographic separation:

Thin layer chromatography: Merck TLC Silica Gel 60 F₂₅₄ aluminium sheets, layer thickness 0.2 mm; Merck TLC Silica Gel 60 RP-18 F₂₅₄S aluminium sheets, layer thickness 0.2 mm.

Column chromatography: Merck Silica Gel 60 F₂₅₄; Merck Silica Gel 60 RP-18 F₂₅₄S

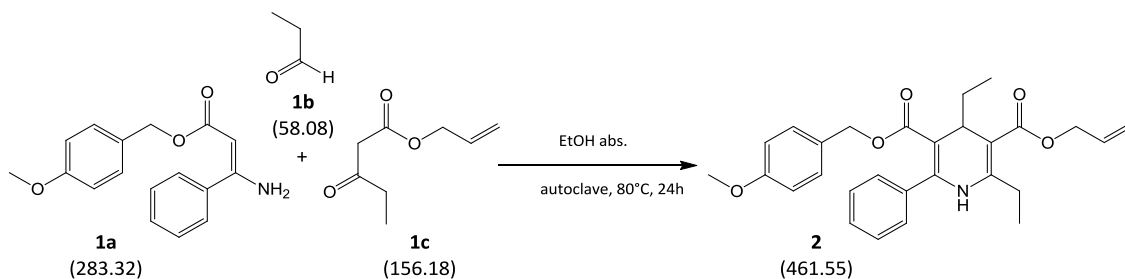
Other equipment:

Melting point determination: Reichert-Kofler hot-stage microscope

CHN analysis: Microanalytic Laboratory, University of Vienna

3.2 Preparation of 4,6-diethyl-2-phenyl-5-thiocarboxy nicotinic acid (7)⁷¹

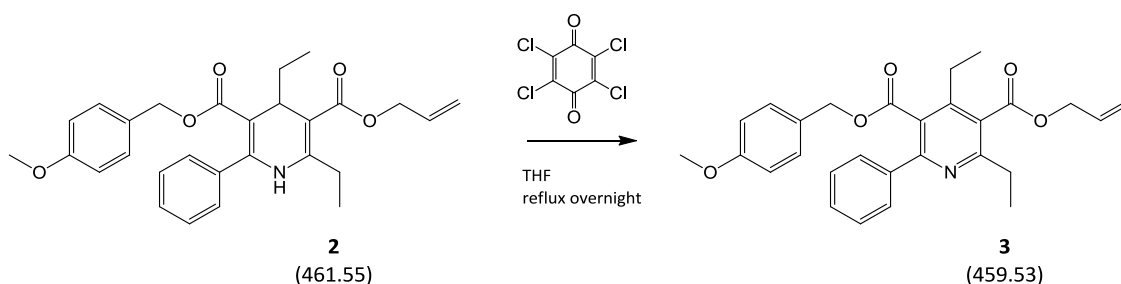
3.2.1 Preparation of 3-allyl 5-(4-methoxybenzyl) 2,4-diethyl-6-phenyl-1,4-dihydropyridine-3,5-dicarboxylate (2)⁷¹



4-Methoxybenzyl-3-amino-3-phenylprop-2-enoate (**1a**) (2.57 g, 9.10 mmol), allyl-3-oxo-pentanoate (**1c**) (1.42 g, 9.10 mmol) and propionaldehyde (**1b**) (0.53 g, 9.10 mmol) are dissolved in 20 ml absolute ethanol and stirred in the autoclave at 80°C for 24 hours. The solvent is removed after cooling down to room temperature and the crude product purified by column chromatography (ligroine/EtOAc 8+2).

Yield: 3.61 g (86 %; yellow-greenish oil).

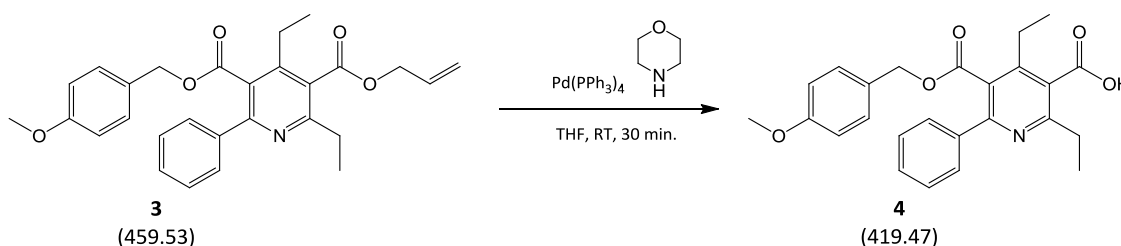
3.2.2 Preparation of 3-allyl 5-(4-methoxybenzyl) 2,4-diethyl-6-phenylpyridine-3,5-dicarboxylate (**3**)⁷¹



3-Allyl 5-(4-methoxybenzyl) 2,4-diethyl-6-phenyl-1,4-dihydropyridine-3,5-dicarboxylate (**2**) (3.61 g, 7.82 mmol) and 2,3,5,6-tetrachloro-1,4-benzoquinone (2.13 g, 8.66 mmol) are dissolved in 250 ml THF and heated under reflux overnight. The reaction mixture is cooled down and the solvent removed under reduced pressure to give the desired pyridine derivative (**3**). The crude product is purified by column chromatography (ligroine/EtOAc 9+1).

Yield: 1.70 g (47 %; orange-brownish oil).

3.2.3 Preparation of 2,4-diethyl-5-[(4-methoxybenzyl)oxy]carbonyl}-6-phenylnicotinic acid (**4**)⁷¹

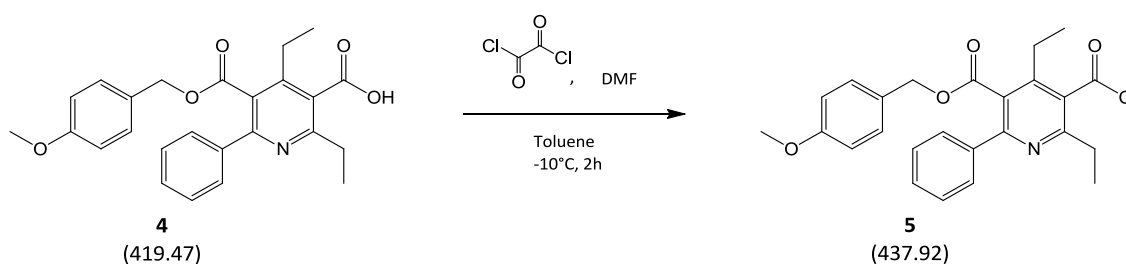


3-Allyl-5-(4-methoxybenzyl)-2,4-diethyl-6-phenylpyridine-3,5-dicarboxylate (**3**) (0.93 g, 2.02 mmol), obtained from the previous step are dissolved in 15 ml THF and then tetrakis(triphenylphosphine)palladium (0.28 g, 0.20 mmol, 10 mol-%) and morpholine (2.11 g, 2.12 ml, 24.21 mmol) are added under argon atmosphere. The mixture is stirred for 30 minutes at room temperature. The solvent is

removed under reduced pressure and the crude product is purified by column chromatography (CH₂Cl₂/CH₃OH 95+5 and EtOAc/MeOH 8+2).

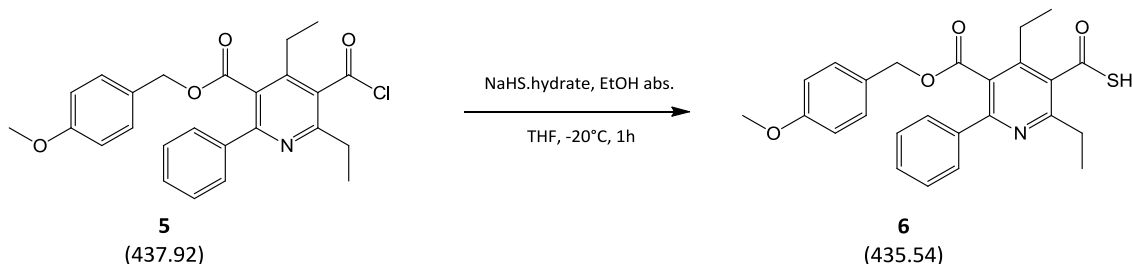
Yield: 806 mg (95 %; yellow crystals, mp. 137-139° C).

3.2.4 Preparation of 4-methoxybenzyl 5-(chlorocarbonyl)-4,6-diethyl-2-phenylnicotinate (**5**)⁷¹



2,4-Diethyl-5-([(4-methoxybenzyl)oxy]carbonyl}-6-phenylnicotinic acid (**4**) (1.00 g, 2.38 mmol) is dissolved in 10 ml dry toluene under inert gas apparatus and cooled down to -20°C. 3 drops of DMF and oxalyl chloride (1 ml) are added drop wise. After reaching room temperature, the mixture is stirred for 1 hour. The solvent is removed under reduced pressure at a temperature below 32°C. The crude product is used immediately for the next reaction step (see 3.2.5).

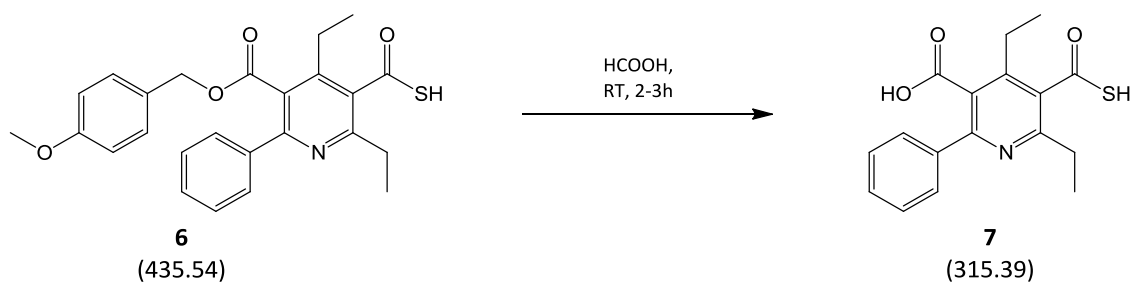
3.2.5 Preparation of 2,4-diethyl-5-[(4-methoxybenzyl)oxy]carbonyl}-6-phenylpyridine-3-carbothioic S-acid (**6**)⁷¹



NaHS.hydrate platelets (0.36 g, 6.45 mmol) are dissolved in 10 ml of absolute ethanol under moderate heating. The solution is cooled down to -20° C. To this solution, the crude 4-methoxybenzyl 5-(chlorocarbonyl)-4,6-diethyl-2-phenylpyridine-3-carboxylate (**5**), obtained from the previous reaction, dissolved in 4 ml of dry THF is added slowly under stirring. After the adding has finished, the reaction mixture is allowed to reach room temperature. The stirring is continued for approximately one hour until the reaction is complete. The orange solution is acidified by 6N HCl solution. The aqueous layer is extracted thoroughly with EtOAc, washed twice with a saturated NaCl solution, and dried over Na₂SO₄. The solvent is removed under reduced pressure and the crude product purified by column chromatography several times (ligroine/EtOAc 7+3).

Yield: 480 mg (46 %) colourless crystals, mp. 59-60° C.

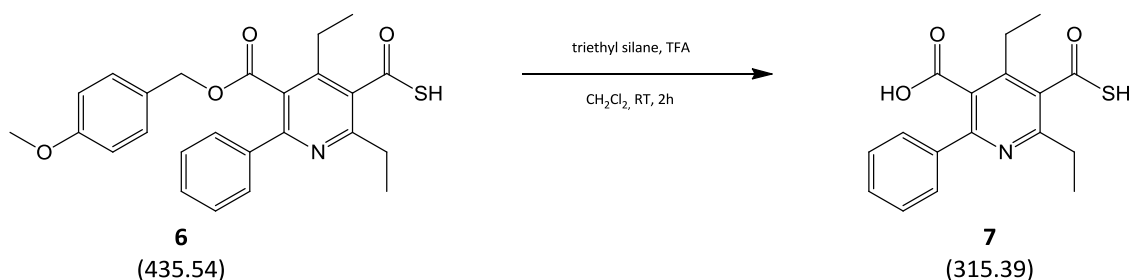
3.2.6 Preparation of 4,6-diethyl-2-phenyl-5-thiocarboxynicotinic acid (SUPPY:o) (**7**)⁷¹



2,4-Diethyl-5-([(4-methoxybenzyl)oxy]carbonyl)-6-phenylpyridine-3-carbothioic S-acid (**6**) (1.00 g, 2.29 mmol) from the previous reaction step is stirred in the presence of 10 ml HCOOH at room temperature for several hours. The solvent is removed under reduced pressure and the crude product is purified by reversed-phase column chromatography (CH₃CN/H₂O mixture 95+5).

Yield: 300 mg (41.5 %; colourless crystals, mp. 191° C).

3.2.6.1 Deprotection with triethyl silane / trifluoroacetic acid

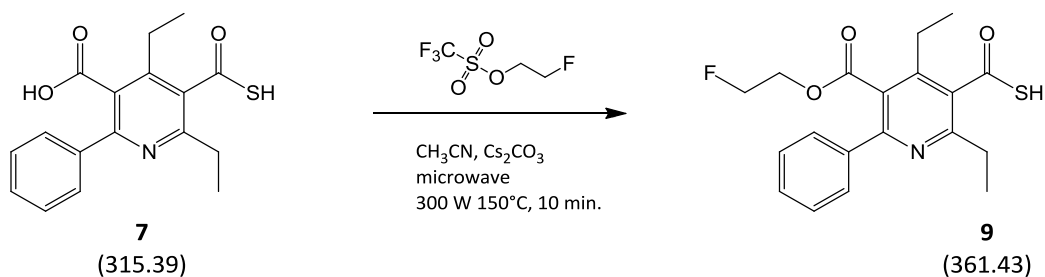


2,4-Diethyl-5-([(4-methoxybenzyl)oxy]carbonyl)-6-phenylpyridine-3-carbothioic S-acid (**6**) (0.10 g, 0.23 mmol) are dissolved in 7 ml of absolute CH_2Cl_2 under argon atmosphere and cooled to 0°C . Triethyl silane (913 μl , 5.74 mmol) and trifluoroacetic acid (2.2 ml, 28.56 mmol) are added slowly. After reaching room temperature, the solution is stirred for further two hours. In the following, the solvent is removed under reduced pressure. The crude product is purified by reversed-phase column chromatography ($\text{CH}_3\text{CN}/\text{H}_2\text{O}$ 95+5).

Yield: 30 mg (41.4 %; colourless crystals, mp. 191°C).

3.3 Preparation of 2-fluoroethyl 4,6-diethyl-5-[(2-fluoroethyl)thio]carbonyl}-2-phenylnicotinate (10)

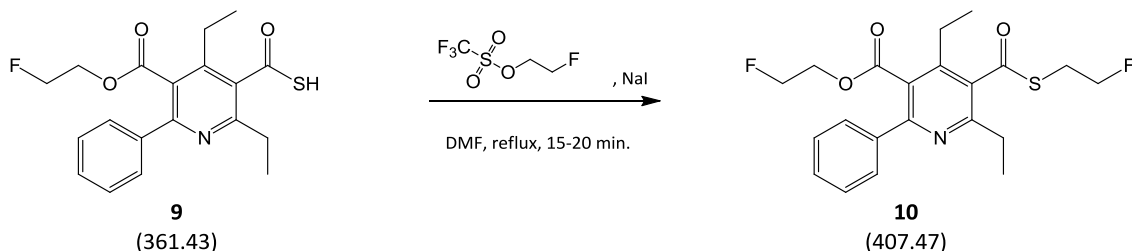
3.3.1 Microwave-assisted preparation of 2,4-diethyl-5-[(2-fluoroethoxy)carbonyl]-6-phenylpyridine-3-carbothioic S-acid (9)⁷¹



4,6-Diethyl-2-phenyl-5-thiocarboxynicotinic acid (**7**) (0.10 g, 0.32 mmol), 2-fluoroethyl trifluoromethanesulfonate (0.14 g, 0.66 mmol) and Cs_2CO_3 (0.22 g, 0.66 mmol) are suspended in 10 ml of CH_3CN . The mixture is heated in the microwave oven at 300 W and 150° C for 10 minutes. The solvent is removed under reduced pressure and the crude product purified by reversed-phase column chromatography (CH_3CN).

Yield: 80 mg (69.2 %; colourless crystals, mp. 45-47° C).

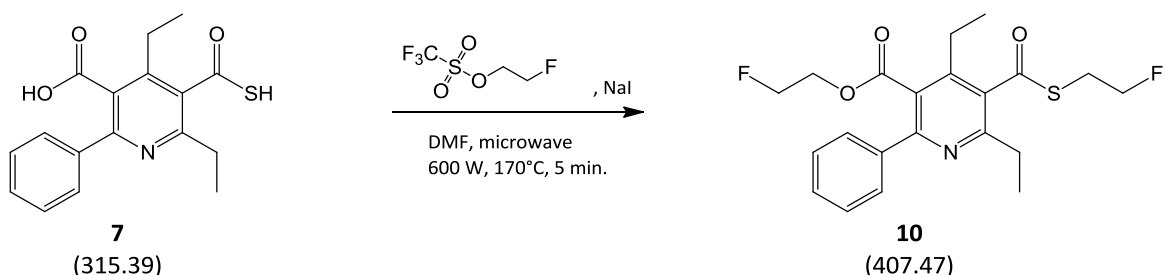
3.3.2 Preparation of 2-fluoroethyl 4,6-diethyl-5-[(2-fluoroethyl)thio]carbonyl}-2-phenylnicotinate (10)



2,4-Diethyl-5-[(2-fluoroethoxy)carbonyl]-6-phenylpyridine-3-carbothioic S-acid (**9**) (0.08 g, 0.22 mmol) from the previous reaction step, NaI (0.33 g, 2.2 mmol) and 2-fluoroethyl trifluoromethanesulfonate (0.12 g, 0.61 mmol) are dissolved in 10 ml of DMF and heated under reflux for 15-20 minutes. The solvent is removed by ball tube distillation and additional drying on a vacuum pump. The crude product is purified by reversed-phase column chromatography ($\text{CH}_3\text{CN}/\text{H}_2\text{O}$ 9+1).

Yield: 51 mg (56.9 %; orange oil).

3.3.3 Microwave assisted single-step preparation of 2-fluoroethyl 4,6-diethyl-5-[(2-fluoroethyl)thio]carbonyl}-2-phenylnicotinate (10)



4,6-Diethyl-2-phenyl-5-thiocarboxynicotinic acid (**7**) (0.073 g, 0.23 mmol), NaI (0.348 g, 2.3 mmol) and 2-fluoroethyl trifluoromethanesulfonate (0.11 g, 0.56 mmol) are dissolved in 10 ml of DMF. The mixture is heated in the microwave

oven at 600 W and 170° C for 5 minutes. The solvent is removed under reduced pressure and the crude product purified by reversed-phase column chromatography (CH₃CN/H₂O 9+1).

Yield: 67 mg (71.5 %; orange oil).

Spectroscopic Data:

¹H NMR (Chart 1.1.) (200 MHz, CDCl₃) δ (ppm): 7.60 (m, 2H), 7.43 (m, 3H), 4.77 (t, 1H, *J* = 5.99 Hz), 4.54 (t, 1H, *J* = 5.94 Hz), 4.42 (m, 1H), 4.34 (q, 1H, *J* = 2.48 Hz), 4.18 (d, 2H, *J* = 5.18 Hz), 3.52 (t, 1H, *J* = 5.99 Hz), 3.41 (t, 1H, *J* = 5.94 Hz), 2.87 (q, 2H, *J* = 7.49 Hz), 2.73 (q, 2H, *J* = 7.53 Hz), 1.34 (t, 3H, *J* = 7.51 Hz), 1.24 (t, 3H, *J* = 7.57 Hz).

¹³C-NMR (Chart 1.2) (50 MHz, CDCl₃) δ (ppm): 194.3, 168.1, 159.6, 157.5, 148.2, 139.6, 132.6, 129.0, 128.4, 128.3, 126.0, 83.0, 82.2, 79.6, 78.8, 64.3, 63.9, 30.2, 29.8, 29.2, 24.2, 15.7, 14.0.

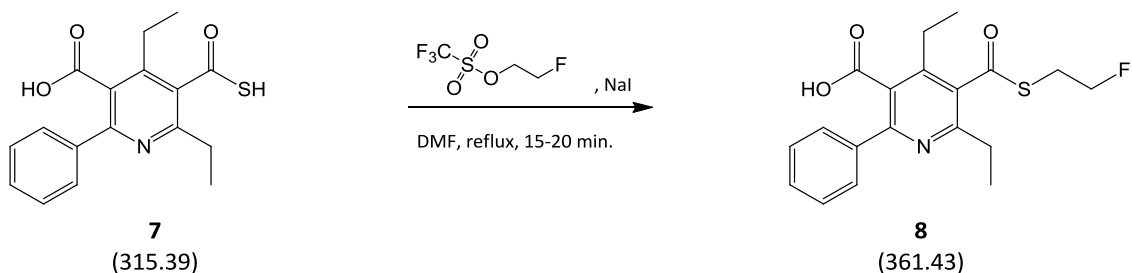
IR (Chart 1.3) (KBr): *ν* (cm⁻¹) 3447, 2977, 2879, 1734, 1675, 1558, 1496

MS (Chart 1.4): *m/z* (%): 407 (M⁺, 0.59), 329 (21), 328 (100), 282 (6), 254 (6), 236 (5), 47 (6).

HRMS (Chart 1.5): *m/z* calculated for C₂₁H₂₃O₃F₂NSH: 408.1445. Found: 408.1450.

3.4 Preparation of methyl 4,6-diethyl-5-[(2-fluoroethyl)thio]carbonyl}-2-phenylnicotinate (**11**)

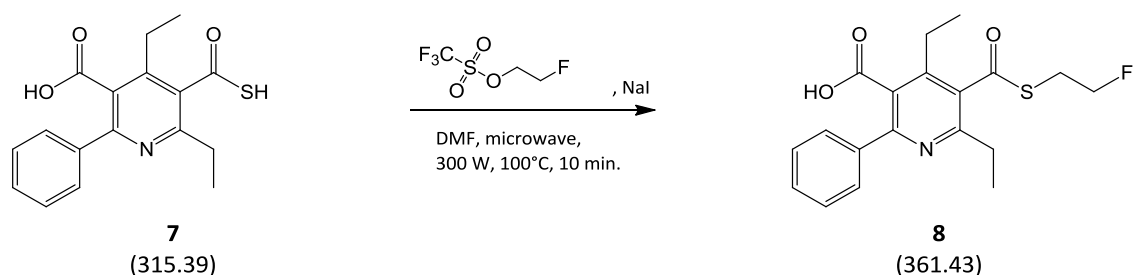
3.4.1 Preparation of 4,6-diethyl-5-[(2-fluoroethyl)thio]carbonyl}-2-phenylnicotinic acid (**8**)



4,6-Diethyl-2-phenyl-5-thiocarboxynicotinic acid (**7**) (0.10 g, 0.32 mmol), NaI (0.476 g, 3.2 mmol) and 2-fluoroethyl trifluoromethanesulfonate (0.14 g, 0.71 mmol) are dissolved in 10 ml of DMF and heated under reflux for 15-20 minutes. The solvent is removed by ball tube distillation. The residue is dissolved in EtOAc and alkalized with a 2N NaOH solution. The organic layer is extracted with water. The aqueous layer is concentrated under reduced pressure, acidified with 2N HCl solution, and thoroughly extracted with EtOAc. The solvent is removed under reduced pressure and the crude product purified by reversed-phase column chromatography (CH₃CN/H₂O 9+1).

Yield: 79 mg (68.3 %; colourless crystals, mp. 195-196° C).

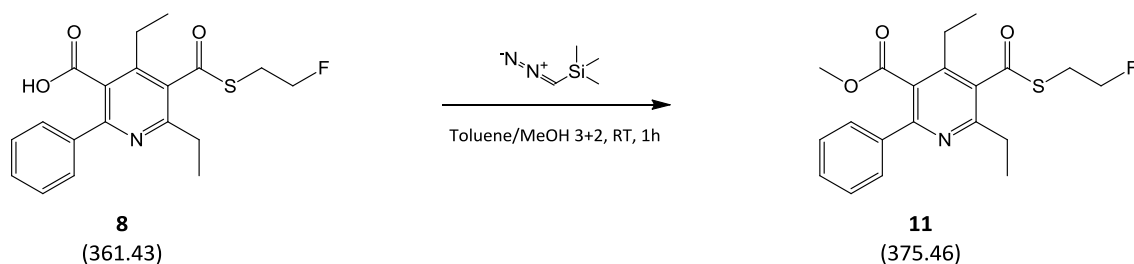
3.4.2 Microwave-assisted preparation of 4,6-diethyl-5-[(2-fluoroethyl)thio]carbonyl}-2-phenylnicotinic acid (**8**)



4,6-Diethyl-2-phenyl-5-thiocarboxynicotinic acid (**7**) (0.05 g, 0.16 mmol), 2-fluoroethyl trifluoromethanesulfonate (0.07 g, 0.36 mmol), and NaI (0.539 g, 3.6 mmol) are dissolved in 10 ml of DMF. The reaction mixture is heated in the microwave oven at 300 W and 100° C for 10 minutes. The solvent is removed by ball tube distillation. The residue is dissolved in EtOAc and alkalized with a 2N NaOH solution. The organic layer is extracted with water. The aqueous layer is concentrated under reduced pressure, acidified with 2N HCl solution, and thoroughly extracted with EtOAc. The solvent is removed under reduced pressure and the crude product purified by reversed-phase column chromatography ($\text{CH}_3\text{CN}/\text{H}_2\text{O}$ 9+1).

Yield: 30 mg (51.9 %; colourless crystals, mp. 195-196 °C).

3.4.3 Preparation of methyl 4,6-diethyl-5-[[2-(2-fluoroethyl)thio]carbonyl]-2-phenylnicotinate (**11**)



4,6-Diethyl-5-[[2-(2-fluoroethyl)thio]carbonyl]-2-phenylnicotinic acid (**8**) (0.079 g, 0.22 mmol) are dissolved in 10 ml of a mixture of toluene/methanol 3+2. (Diazomethyl)trimethylsilane solution in hexane 2.0M (0.16 ml, 0.33 mmol) is added under continuous stirring until the yellow colour persists. The stirring is continued for 30 minutes. If the reaction is incomplete, additional (diazomethyl)trimethylsilane solution in hexane 2.0M is added (0.16 ml, 0.33 mmol) and stirring continued for further 30 minutes. After an overall stirring of one hour the reaction is stopped. The solvent is removed under reduced pressure and the crude product purified by reversed-phase column chromatography (acetonitrile/water 9+1).

Yield: 73 mg (88.4 %; colourless crystals, mp. 55-57° C).

Spectroscopic Data:

¹H NMR (Chart 2.1) (200 MHz, CDCl₃) δ (ppm): 7.59 (q, 2H, *J* = 2.86 Hz), 7.44 (m, 3H), 4.77 (t, 1H, *J* = 5.93 Hz), 4.54 (t, 1H, *J* = 5.99 Hz), 3.63 (s, 3 H), 3.51 (t, 1H, *J* = 5.99 Hz), 3.40 (t, 1H, *J* = 6.00 Hz), 2.86 (q, 2H, *J* = 7.50 Hz), 2.71 (q, 2H, *J* = 7.58 Hz), 1.34 (t, 3H, *J* = 7.45 Hz), 1.22 (t, 3H, *J* = 7.57 Hz).

¹³C-NMR (Chart 2.2) (50 MHz, CDCl₃) δ (ppm): 194.5, 168.9, 159.4, 157.1, 148.1, 139.6, 132.6, 128.4, 128.2, 126.3, 83.0, 79.6, 52.3, 30.2, 29.8, 29.2, 24.2, 15.7, 13.9.

IR (Chart 2.3) (KBr): *ν* (cm⁻¹) 3427, 2975, 2884, 1725, 1653, 1558, 1495.

MS (Chart 2.4): *m/z* (%): 375 (M⁺, 0.90), 297 (18), 296 (100), 264 (5), 236 (8).

HRMS (Chart 2.5): *m/z* calculated for C₂₀H₂₂O₃FNSH: 376.1383. Found: 376.1392.

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5. APPENDIX

5.1 Spectra

Chart 1.1

2-Fluoroethyl 4,6-diethyl-5-{-[(2-fluoroethyl)thio]carbonyl}-2-phenylnicotinate in CDCl₃

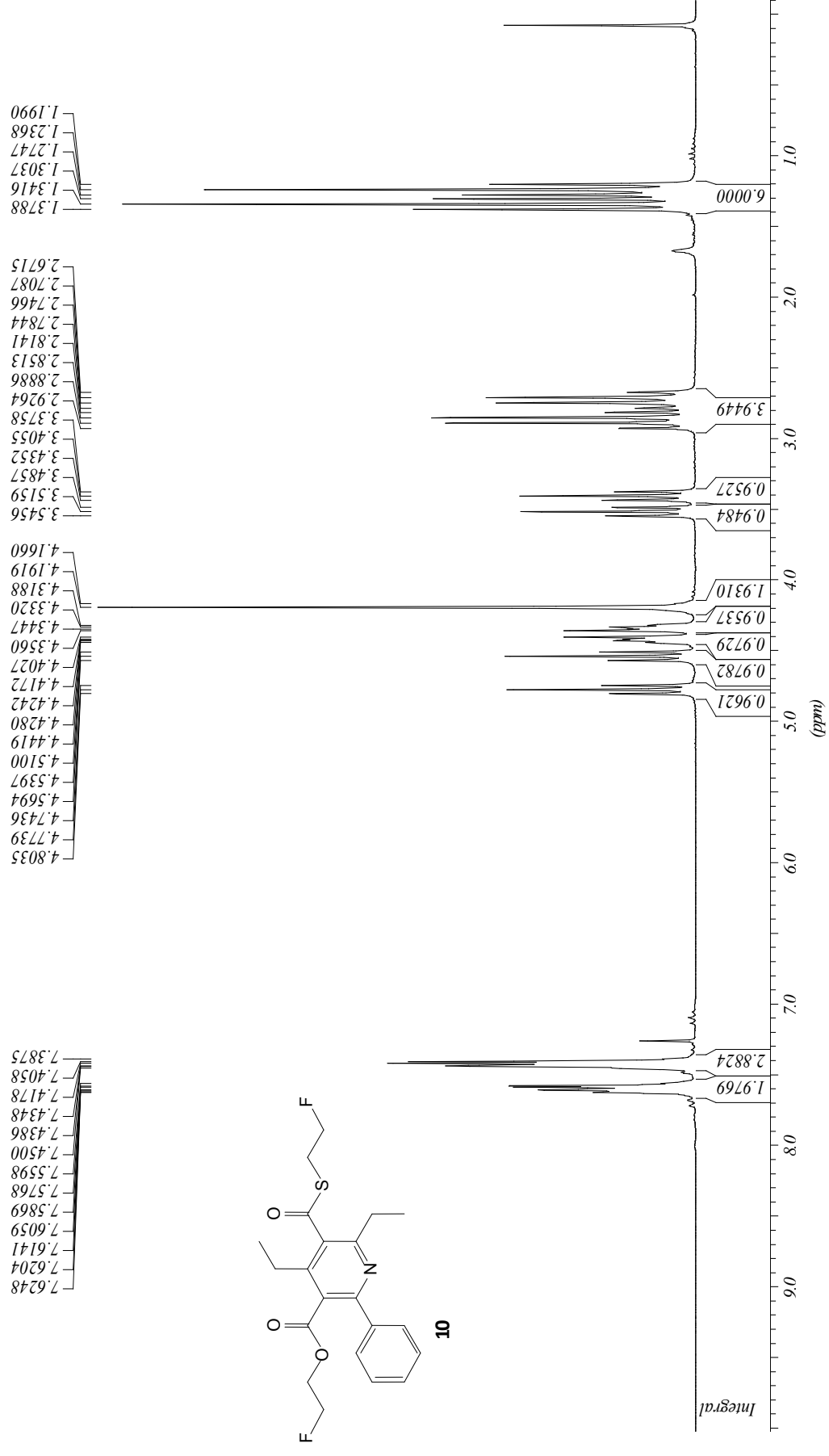


Chart 1.2

2-Fluoroethyl 4,6-diethyl-5-{[(2-fluoroethyl)thio]carbonyl}-2-phenylnicotinate in CDCl₃

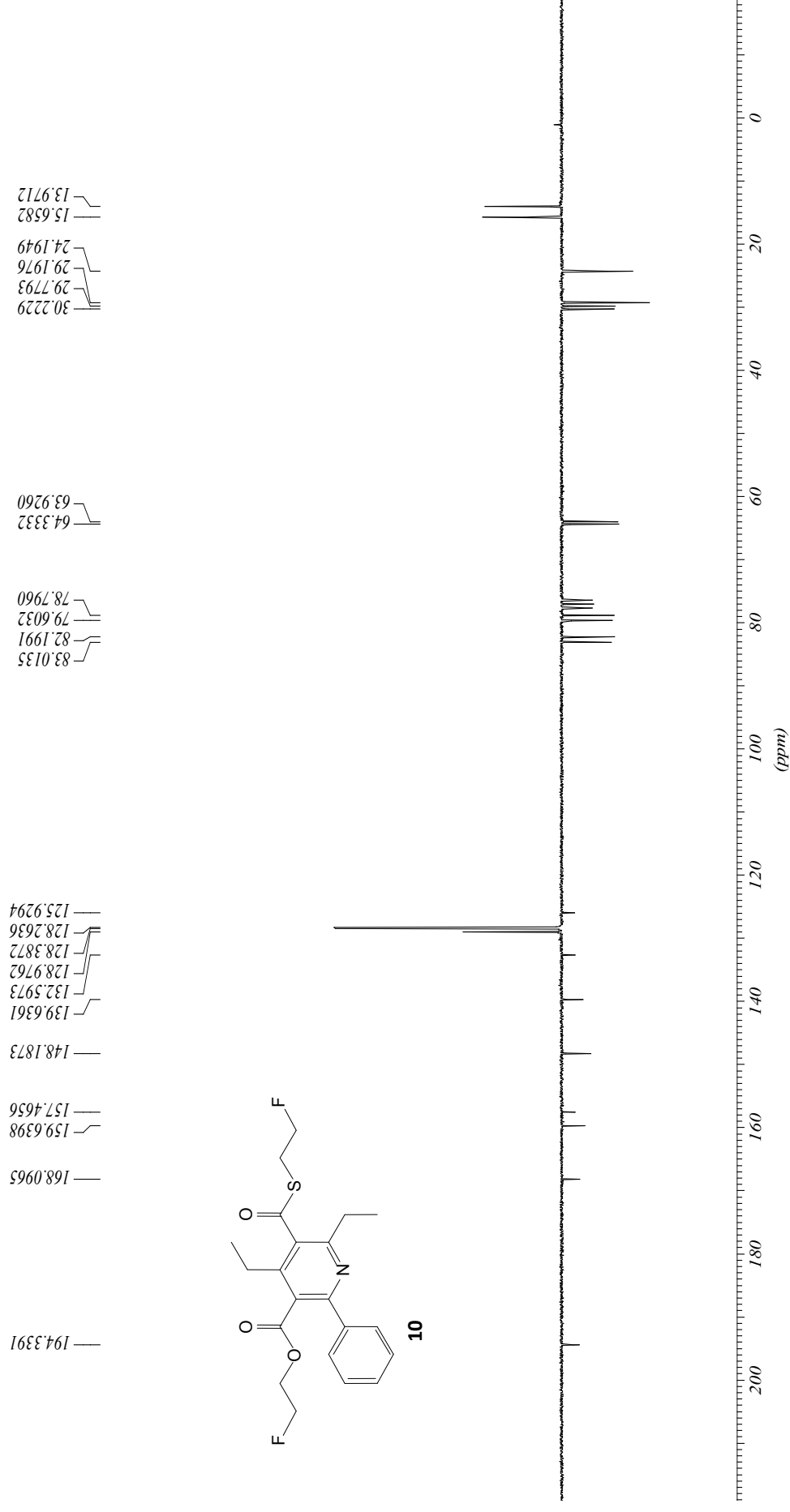


Chart 1.3 2-Fluoroethyl 4,6-diethyl-5-{-[(2-fluoroethyl)thio]carbonyl}-2-phenylnicotinate

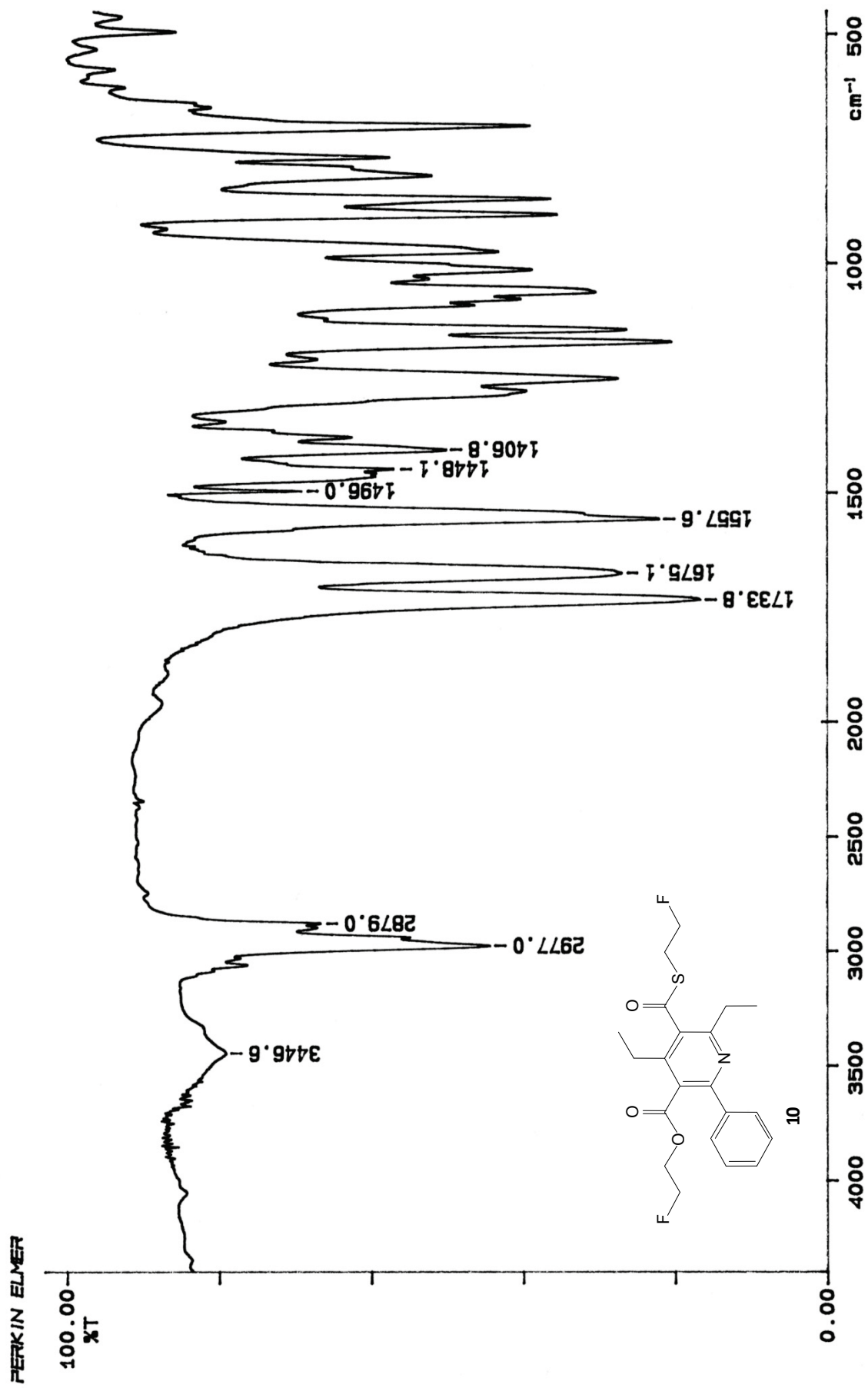


Chart 1.4

2-Fluoroethyl 4,6-diethyl-5-[[[(2-fluoroethyl)thio]carbonyl}-2-phenyl]nicotinate

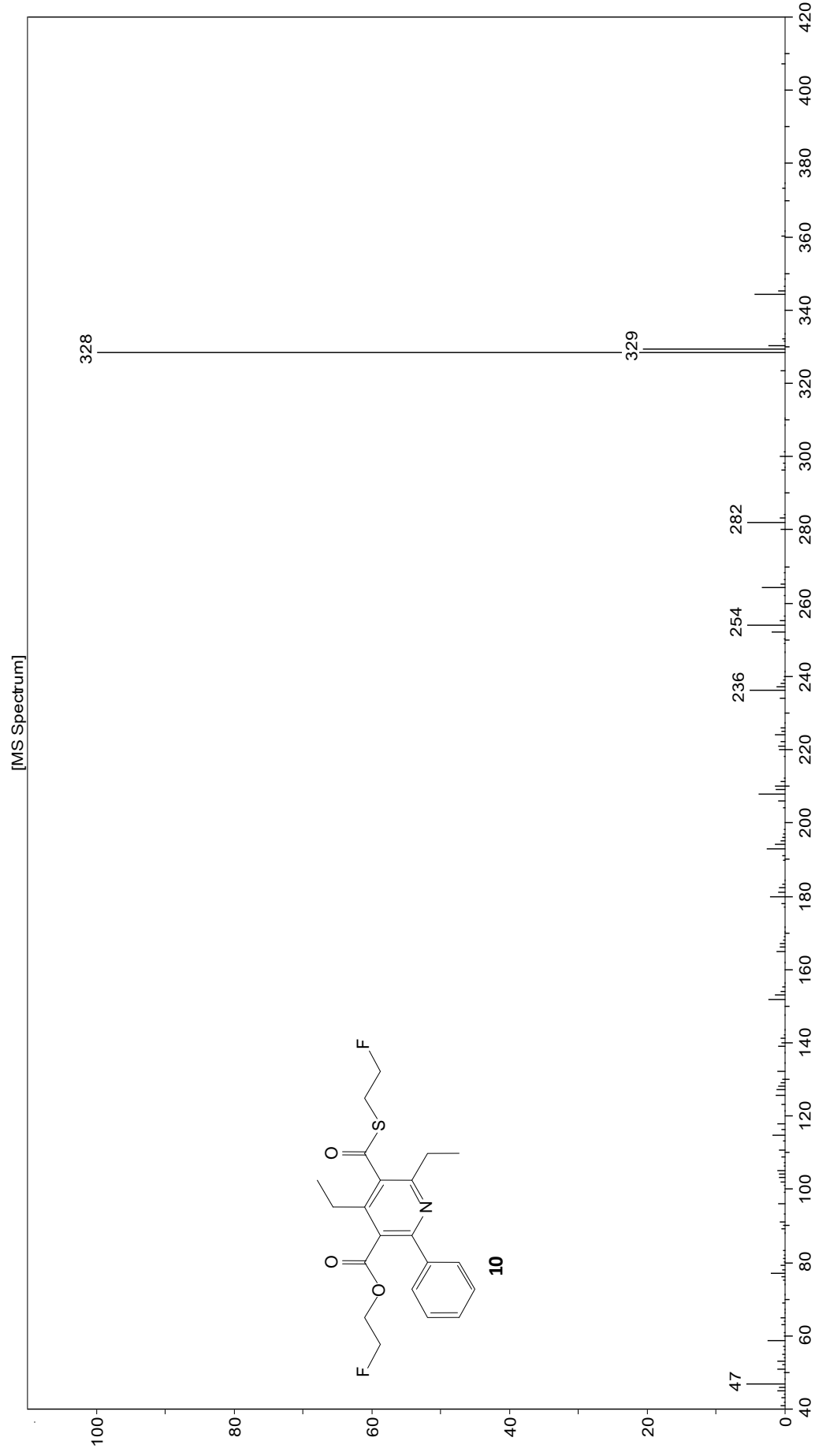


Chart 2.1

Methyl 4,6-diethyl-5-{-[(2-fluoroethyl)thio]carbonyl}-2-phenylnicotinate in CDCl₃

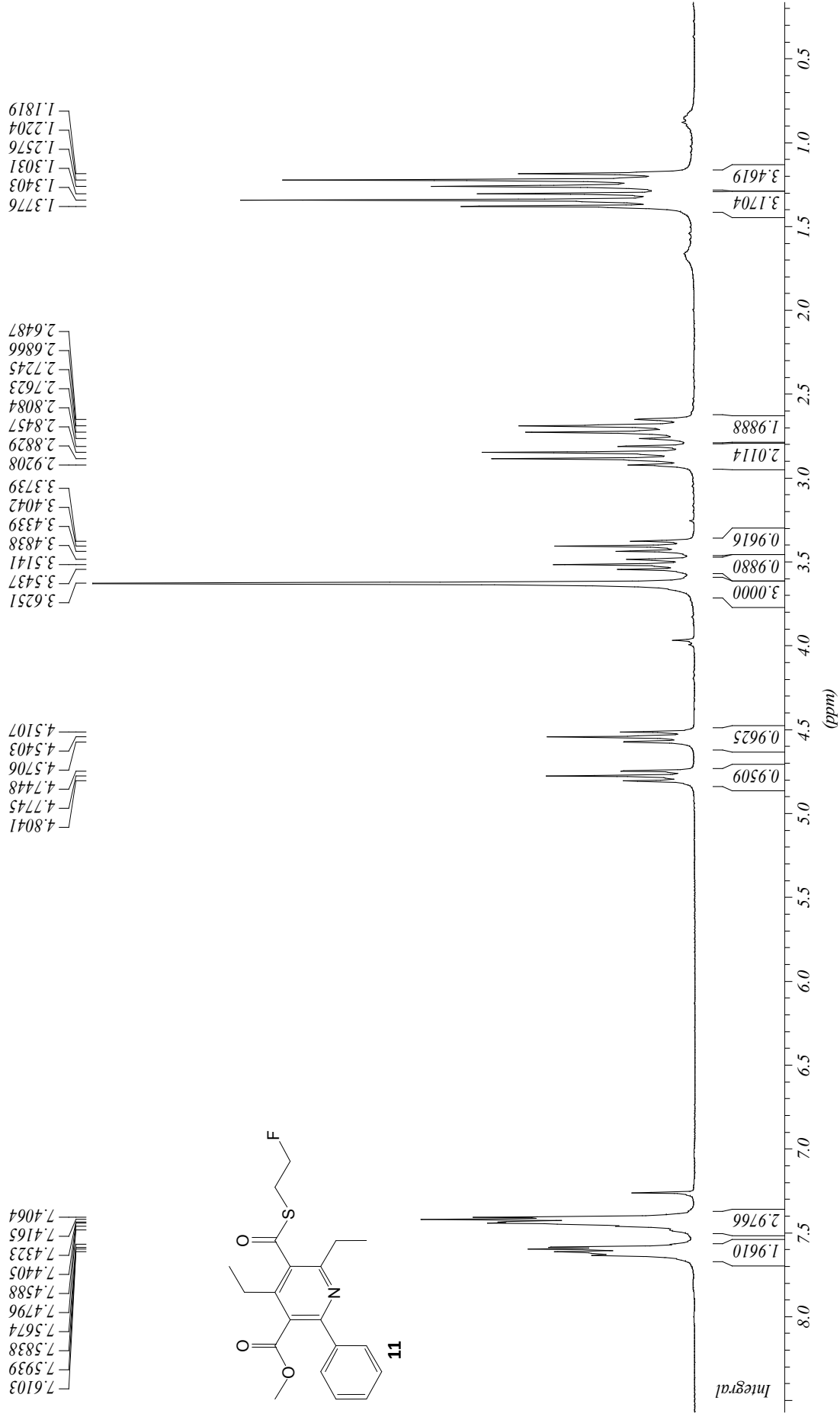


Chart 2.2

Methyl 4,6-diethyl-5-[[{(2-fluoroethyl)thio]carbonyl}-2-phenyl]nicotinate in CDCl₃

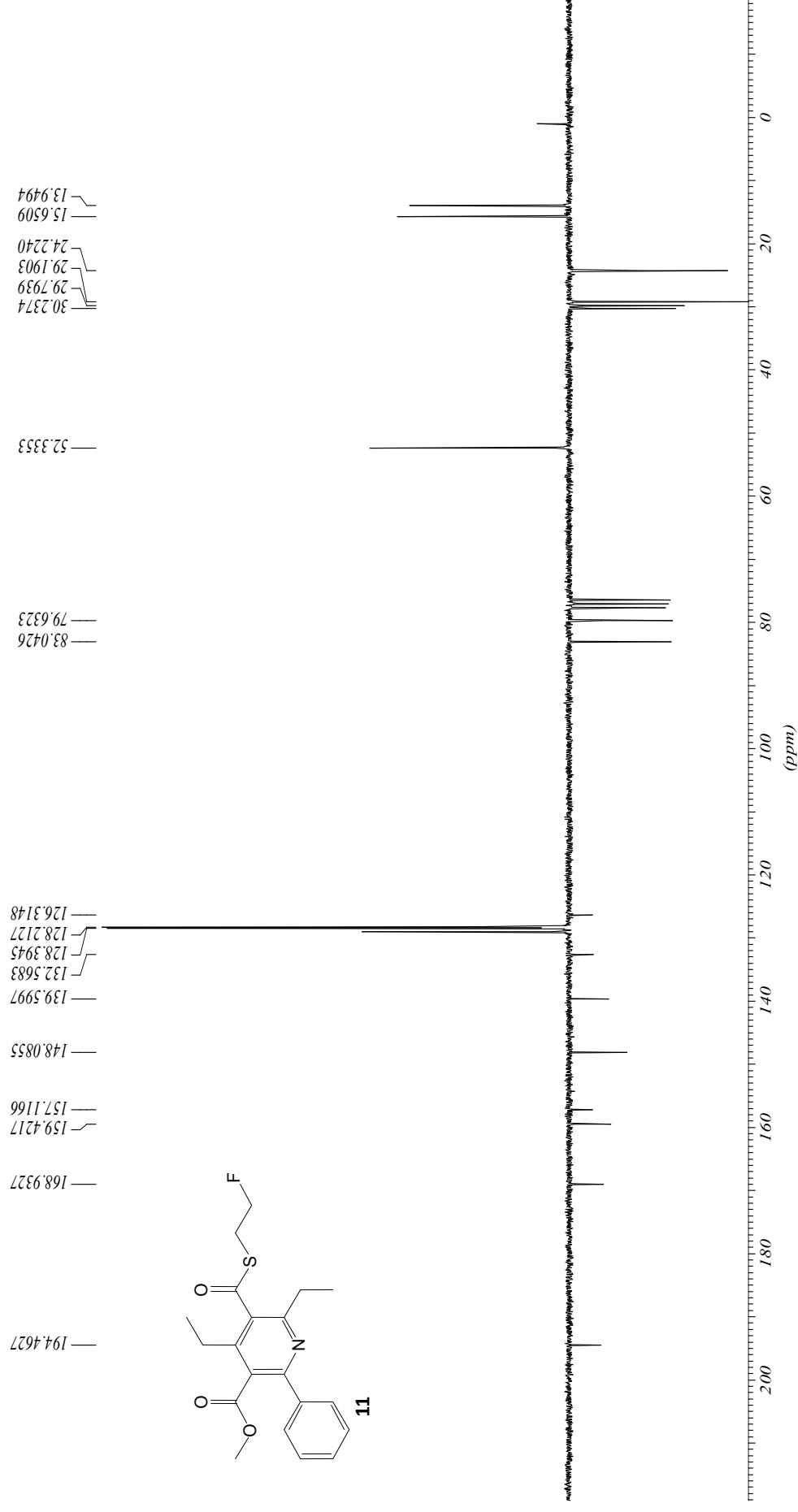


Chart 2.3 Methyl 4,6-diethyl-5-{-[(2-fluoroethyl)thio]carbonyl}-2-phenylnicotinate

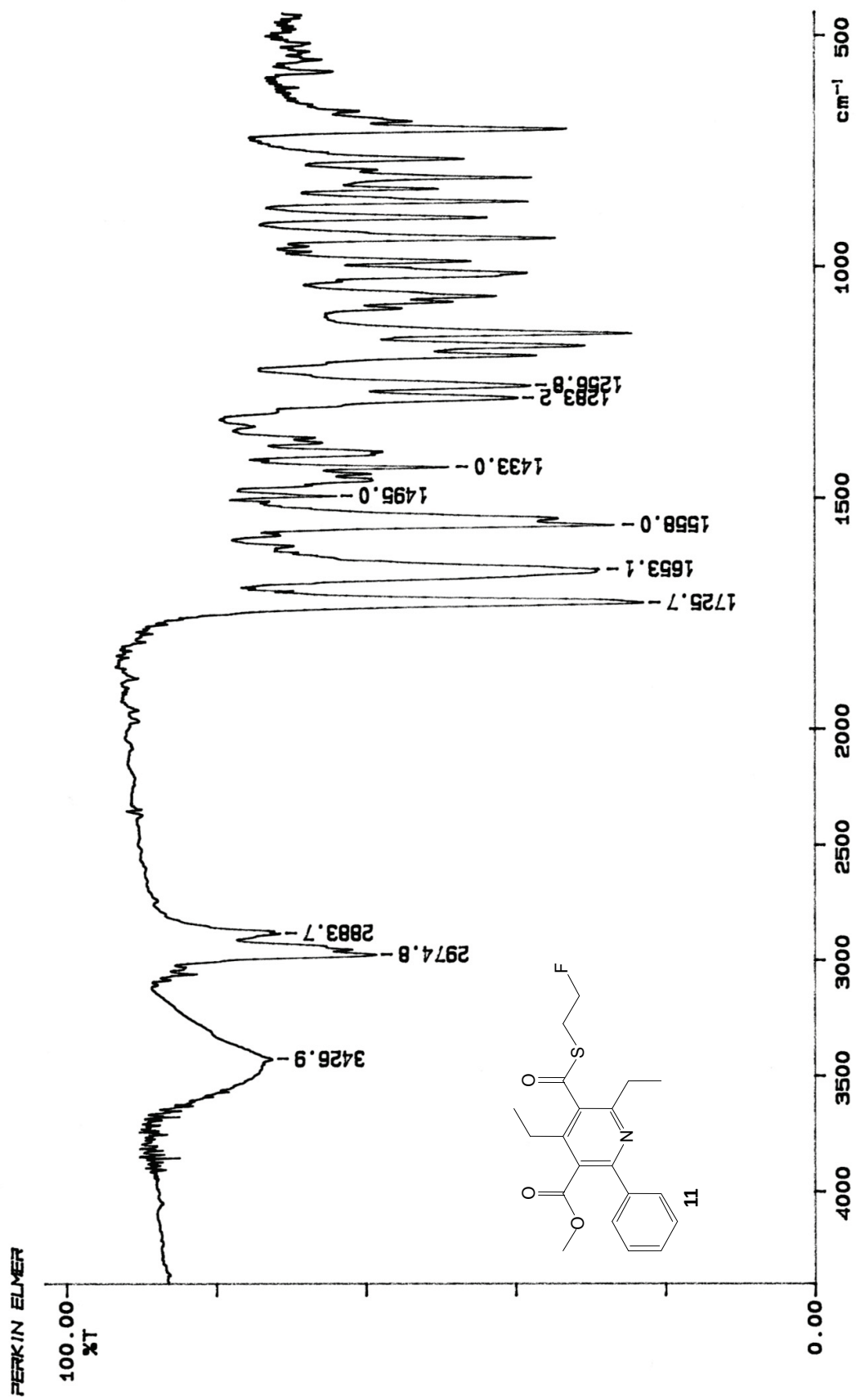
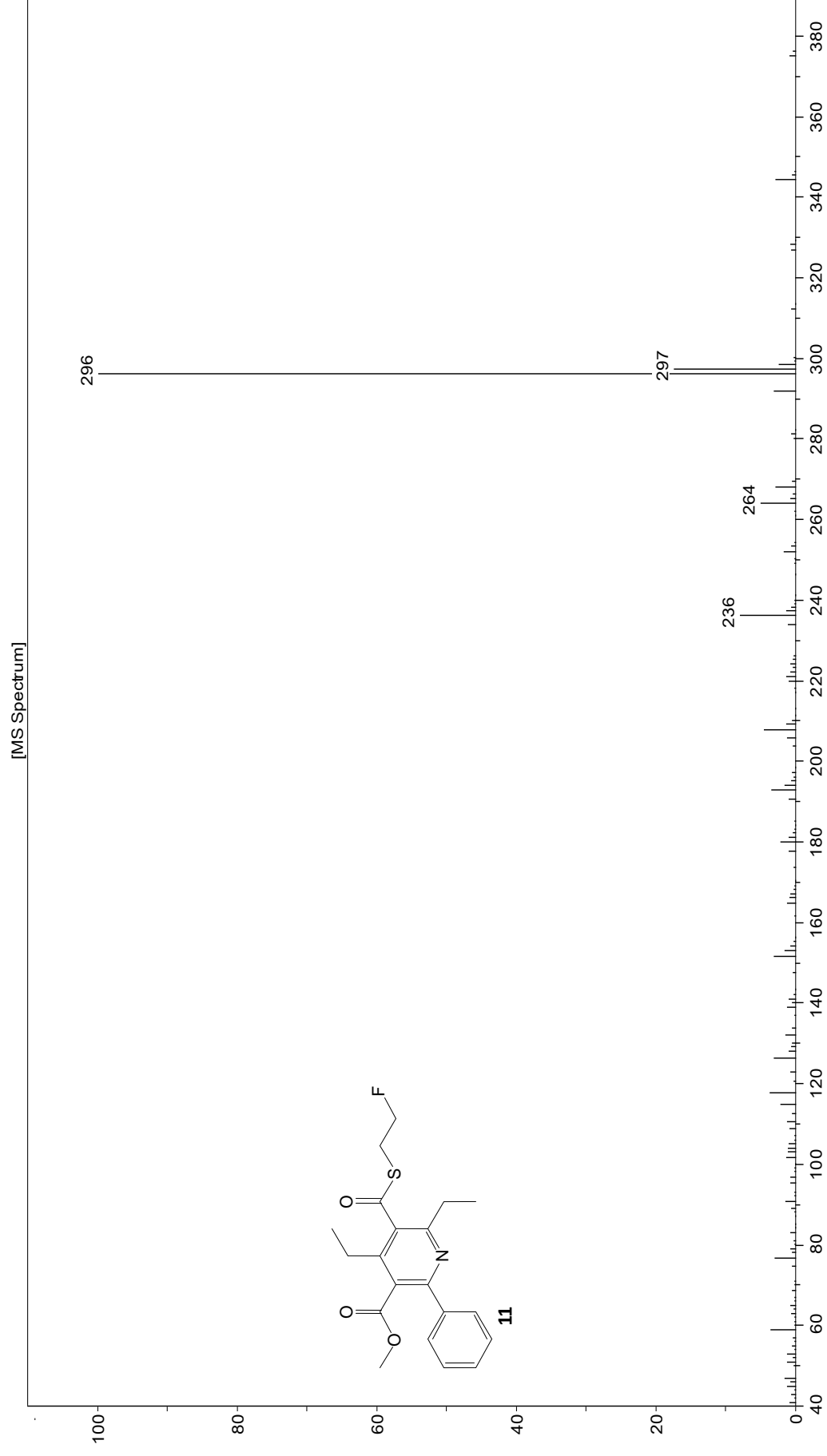


Chart 2.4

Methyl 4,6-diethyl-5-{{[(2-fluoroethyl)thio]carbonyl}}-2-phenylnicotinate



5.2 Abstract

In the past 3 decades, the development of positron emission tomography (PET) has made great progress. It has become a powerful imaging technique for medical diagnostic as well as answering many questions of physiological, biochemical or pharmacokinetic interest. Apart from the technical evolution of the PET scanners, the development of new tracer molecules was a prerequisite necessary for the success of this technique.

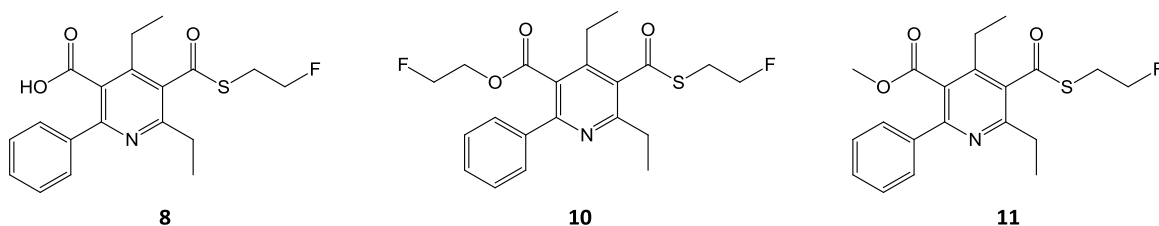
Among others, the adenosine A₃ receptor and its properties has become of major interest since many of its biological functions remained unclear. Subsequently, the preparation of numerous possible PET tracer molecules began.

Eventually, a series of suitable PET antagonists, comprising a sulfanyl-phenyl-pyridine structure, were developed.

In the submitted thesis the preparation of two novel adenosine-A₃-receptor antagonists are described.

They are 2-fluoroethyl 4,6-diethyl-5-[[2-(2-fluoroethyl)thio]carbonyl]-2-phenylpyridine-3-carboxylate (**10**) and methyl 4,6-diethyl-5-[[2-(2-fluoroethyl)thio] carbonyl]-2-phenylpyridine-3-carboxylate (**11**).

Another task was a new synthesis pathway to obtain 4,6-diethyl-5-[[2-(2-fluoroethyl)thio]carbonyl]-2-phenylpyridine-3-carboxylic acid (**8**).



These three molecules should serve as HPLC reference compounds and need further investigation regarding their receptor binding qualities.

5.3 Zusammenfassung

In den vergangenen 3 Jahrzehnten hat die Entwicklung der Positronen-Emissions-Tomographie große Fortschritte erzielt. Sie wurde ein wichtiges bildgebendes Verfahren, sowohl für die medizinische Diagnostik als auch die Forschung. Sie ermöglicht die Beantwortung vieler Fragestellungen in der Physiologie, Biochemie und der Pharmakokinetik.

Abgesehen von der technischen Evolution der PET-Scanner selbst, war die Entwicklung neuer Tracer eine Voraussetzung für den Erfolg der Methode.

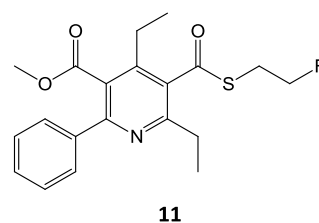
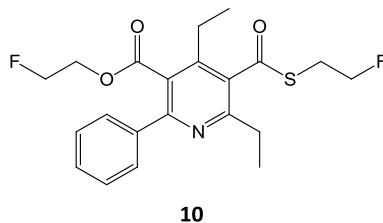
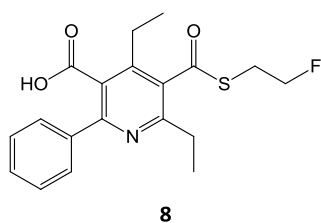
Neben anderen wurde der Adenosin-A₃-Rezeptor und dessen Eigenschaften zum Gegenstand der Forschung, da viele seiner biologischen Funktionen bis dato ungeklärt waren. Dies führte zur Entwicklung einer Reihe potenzieller PET-Tracer.

Schließlich wurde eine Serie geeigneter PET-Antagonisten, bestehend aus einem Sulfanyl-phenyl-pyridin-Grundgerüst, entwickelt.

In der vorgelegten Arbeit wird die Synthese zweier Adenosin-A₃-Rezeptor-Antagonisten beschrieben.

Die zwei Moleküle sind 2-Fluorethyl 4,6-diethyl-5-[[[(2-fluorethyl)thio] carbonyl]-2-phenylnicotinat (**10**) und Methyl 4,6-diethyl-5-[[[(2-fluorethyl)thio]carbonyl]-2-phenylnicotinat (**11**).

Die zweite Aufgabe war ein neuer Syntheseweg für 4,6-diethyl-5-[[[(2-fluorethyl)thio]carbonyl]-2-phenylnicotinsäure (**8**).



Diese 3 Moleküle sollen als HPLC Referenzsubstanzen dienen und werden in Zukunft möglicherweise auf ihre Rezeptorbindungseigenschaften getestet.

5.4 Curriculum Vitae

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