SCIENTIFIC DISCUSSION

This module reflects the initial scientific discussion and scientific discussion on procedures, which have been finalised before 1 June 2003. For scientific information on procedures after this date please refer to module 8B.

1. Introduction

NeoSpect is a diagnostic medicinal product containing the active substance depreotide INN as the trifluoroacetate salt indicated for the scintigraphic imaging of suspected malignant tumours in the lung, in combination with a CT scan or chest X-ray, in patients with solitary pulmonary nodules.

NeoSpect is presented as a powder for solution for injection, as a kit for diagnostic use after reconstitution/radiolabelling with sodium pertechnetate (^{99m}Tc) injection. On reconstitution, technetium (^{99m}Tc) depreotide is formed, which contains within the cyclic hexapeptide domain of the peptide component, a pharmacophore which binds to the somatostatin receptor (SSTR) of tumour cells.

Radiotracers and nuclear medicine technology may be used to detect many cancers in their early stages by recognition of tumour-specific binding sites. Several types of tumour have been identified as exhibiting high expression of receptors for somatostatin (SST). The SST receptor is a membrane-adapted glycoprotein that appears to be present in a significant number of human malignancies including both small cell lung cancer (SCLC) and non-small cell lung cancer (NSCLC). This hyper-expression by tumour cells should allow differentiation between normal and cancerous tissue. After specific binding by the peptide component of ^{99m}Tc-labelled NeoSpect, selective imaging can in theory be achieved in the standard way by means of a gamma camera, the principle instrument currently in use for imaging in nuclear medicine.

2. Chemical, pharmaceutical and biological aspects

Composition

NeoSpect is presented as a sterile, freeze-dried powder of depreotide trifluoroacetate (TFA) (47 µg) in 5ml Type I (Ph.Eur) glass vials sealed with grey, siliconised, butyl rubber stoppers. The product also contains: sodium α-D-glucoheptonate dihydrate as a bulking agent and aid in radiolabelling; stannous chloride dihydrate as a reducing agent; disodium edetate as an aid in radiolabelling; hydrochloric acid and sodium hydroxide for pH adjustment.

Active substance

Depreotide (INN) is a synthetic 10 amino acid peptide with the chemical name cyclo(-L-homocysteinyl-N-methyl-L-phenylalanyl-L-tyrosyl-D-tryptophyl-L-lysyl-L-valyl), (1→1')-sulfide with 3[(mercaptoacetyl)amino]-L-alanyl-L-lysyl-L-cysteinyl-L-lysine amide, trifluoroacetate salt. Its structure is:

S-CH_2-CO-β-Dap-Lys-Cys-Lys-NH_2, TFA salt

Cyclo(-homocys-N-Me-Phe-Tyr-D-Trp-Lys-Val)

Synthesis of depreotide involves solid-state synthesis of two intermediate peptides on resins. Each bound peptide is then cleaved from the resin, deprotected and purified by preparative HPLC. The two peptide intermediates (P334 and P802), which are checked for identity and purity, are then coupled and the resulting peptide is deprotected to produce depreotide. Depreotide is purified by preparative HPLC and lyophilised to produce depreotide trifluoroacetate.

Proof of structure has been demonstrated by mass spectrometry (fast atom bombardment and electrospray ionisation), H-NMR spectroscopy (which confirms the assigned amino acid sequence and structure), amino acid analysis (a standard technique used for analysis of peptides), elemental analysis and peptide optical purity.

The specification proposed for the active substance, depreotide trifluoroacetate, is comprehensive and has been revised during the evaluation procedure. Several identity tests are required to conclusively demonstrate the identity of the active substance. The specification also now includes tests and limits (which have been justified) for: content and purity of peptide; depreotide content; trifluoroacetate content; known and unknown peptide impurities; amino acid enantiomeric impurities; other impurities; residual solvents (acetonitrile, dichloromethane, diethyl ether, triethylamine, trisopropylsilane and total); water content; mass balance; bioburden limit, and endotoxin content. Further information was provided on the toxicity of trisopropylsilane in order to justify the limit proposed. Batch analyses data have been provided for three batches of the active substance and these demonstrate compliance with the revised specification.

A discussion of potential impurities is included. Reference is also made to published literature. The possible formation of diastereoisomers, both deletion and truncated sequences, and incomplete deprotection is discussed. The formation of diastereoisomers of depreotide during synthesis can result from using optically impure amino acid derivatives. The starting materials are therefore checked for optical purity. Other supporting data have indicated that the method of synthesis does not cause racemization of the amino acids. Racemization only becomes a problem in prolonged slow coupling reactions but in the case of depreotide none of the reactions is prolonged or slow. Therefore the potential for racemization is stated to be minimal. A test for optical rotation is therefore not included in the drug substance specification.

Impurities found include:

- Two primary synthetic impurities, impurity 'o' and impurity 'p', they are limited in the specification at levels which have been justified by toxicology studies.
- Six primary degradation products were identified and coded as 'd', 'e', 'f', 'g', 'h' and P1066. Degradants 'd', 'e', 'f', 'g', have been ascribed to oxygenation products. They are controlled individually in the specification. The degradant 'h' has been ascribed to a base hydrolysis product of depreotide and the limit of 1.0% is justified and acceptable. Tentative proposals to assign a structure to 'h' have been attempted, but as this impurity is only present in small amounts in thermally degraded or base hydrolysed samples, further determination of its structure and/or identity is not possible. The primary degradation product in depreotide is the disulfide dimer P1066. It has been characterised by MS, HPLC, H-NMR and arises from oxidation of depreotide, which probably occurs between the time of preparative HPLC purification and lyophilisation. A limit for impurity P1066 of < 1.5% is proposed and justified.
- Additional impurities/degradants found are 'k', 'q', 'r', and 'u'.

The proposed limit of NMT 0.2% for 'other greatest single unknown' impurity is supported by the batch data presented.

Batch analyses data were presented from one commercial scale batch (7g) and three pilot scale batches (2g) used for clinical studies. All pilot batches met the initially proposed specification. As the specification has been revised during the course of the procedure, a follow-up measure is included that further batch analyses data be provided from future commercial scale batches in order to fully justify the amended specification now agreed.

The stability of the active substance has been investigated (using one commercial and five pilot scale batches) at < -10°C with accelerated studies at 5°C ± 3°C. Stress conditions included exposure to acid and base, light, peroxide treatment and elevated temperatures. Results of photostability studies indicated that the drug substance must be stored protected from light. The proposed storage statement of '≤-10°C protected from light' is justified, as the peptide purity of all batches monitored indicated a significant reduction in peptide purity after 6 months when drug was stored at 5°C ± 3°C. A commitment was given to provide stability data for a further two commercial scale batches and to place one commercial scale batch on stability trial yearly thereafter. The proposed retest period for the drug substance is 24 months with retesting for purity immediately before use after 12 months and this has now been justified, (with the follow-up measure described above).

Other ingredients

The excipients stannous chloride dihydrate, disodium edetate, concentrated hydrochloric acid and sodium hydroxide comply with the corresponding current specifications of the PhEur. Water for

injections used as a solvent in the manufacture of the product prior to its freeze drying also conforms to the current PhEur monograph.

Non-pharmacopoeial excipients are sodium α-D-glucoheptonate dihydrate and nitrogen (which is used as an inert atmosphere in the vials). Their quality is adequately controlled by the in house specifications.

Product development and finished product

The formulation used in the pivotal clinical trials carried a nominal label claim of 50 micrograms of active substance per vial, instead of 47 micrograms which is currently proposed for marketing. Earlier trials used a formulation which was modified in 1995 to improve radiochemical purity, by reducing the quantity of sodium α-D glucoheptonate dihydrate from 50 mg to 5 mg per vial and adding sodium edetate at 100 µg per vial. It should also be noted that clinical trial batches involved the earlier reconstitution process of incubation of the reconstituted solution at ambient room temperature. In 1996 the procedure was modified to incubate the reconstituted product in boiling water, which reduced by 3-5% the level of Tc non-mobile species (degradation products).

The optimal concentration of depreotide before reconstitution was investigated. It was shown that lyophilised depreotide formulated at a content in the region of 50 µg/vial showed the best radiochemical purity over 6.5 hours post-reconstitution.

The choice and characteristics of the chosen excipients are satisfactorily shown to be appropriate for their intended use. Sodium α -D glucoheptonate dihydrate and sodium edetate serve as aids in radio-labelling. As a chelating agent, edetate may compete with depreotide for technetium, leading to a poor radiochemical yield of technetium depreotide. Its effect on radiochemical purity (RCP) and stability was assessed and use of 100 µg was shown to be most suitable. Stannous chloride dihydrate reduces ^{99m}Tc from the 7+ oxidation state in pertechnetate to the 5+ oxidation state favourable for binding to nitrogen and sulphur atoms. The quantity of this ingredient is crucial in reducing pertechnetate, leading to efficient formation of technetium depreotide. A level of 50 µg vial was originally chosen as being the most suitable, although this has been revised during the evaluation process (see below).

Presentation of the product in a ready-to-use solution form is not feasible due to its poor aqueous stability. Given the heat labile nature of the drug substance (a peptide), the product is sterilised by filtration, filled aseptically (through a 0.22 µm filter with relevant control of bioburden and bubble point filter integrity testing) into pre-sterilised vials and then lyophilised using a well defined and validated cycle. The bulk solution sterile filtration process results in some binding of depreotide to the filters and as a result of this either the label claim had to be reduced (from the 50 µg/vial originally proposed) or a manufacturing overage added. The applicant chose the first option, reducing the label claim to 47 µg/vial. Given that the clinical studies were all conducted with product containing this quantity and in effect nothing has changed except the nominal expression of content, this is without any material effect on the clinical (diagnostic) efficacy or safety of the product.

Depreotide injection is reconstituted with sodium pertechnetate (^{99m}Tc) injection containing up to 50mCi (1870MBq) of technetium-99m activity.

The radiochemical purity (RCP) of only 80% (by HPLC) does not adversely affect the biological performance of ^{99m}Tc-depreotide. Preparations of 80% and 95% RCP were shown to have equivalent tumour targeting in tumour bearing nude mice and receptor-binding values.

The labelling of depreotide injection with ^{99m}Tc is a critical process. The reconstituted product must be incubated in a boiling water bath or heat block with sodium pertechnetate (^{99m}Tc) injection in normal saline for 10 minutes in order to optimize the formation of technetium (^{99m}Tc) depreotide injection. Development of the reconstitution process and its likely effect on the radiochemical purity and peptide composition have been investigated with respect to incubation time, incubation temperature, reconstitution volume, activity level stopper content and dilution. Findings regarding storage of the reconstituted product showed that the vials should be stored upright.

Performing the labelling with sodium pertechnetate eluates from five European and three US generator manufacturers have tested robustness of the product in clinical use. All were found to be acceptable.

The radiochemical purity of the reconstituted product stored in a polypropylene syringes for up to five hours was found to be satisfactory (80% maintained over the five hours). No appreciable adsorption of the drug substance to the syringe was found after 5 hours of storage.

The reconstituted product (technetium) ^{99m}Tc-depreotide is shown to be present in two isomeric forms in technetium depreotide injection, as the "*syn*" and "*anti*" isomers. It was found that the ratio was consistent from lot to lot and from preparation to preparation. Satisfactory diagrams of structures are also provided.

The specifications for the finished product have been refined during the review process and are now satisfactory to control the quality of this product. Separate specifications are used for the kit and for the reconstituted solution for administration.

For the kit - The identification testing of the peptide cation has been improved by the addition of an HPLC test to the MS method. Likewise, an identity test (^{19}F NMR) has been added for the trifluoroacetate anion. The limits for the content of depreotide (base) are 47 µg/vial ± 7.5% (43 to 50 µg/vial), although this will be tightened further if data from future batches permits. The limits for the excipients, and likewise for the impurities/degradation products have been tightened and justified during the evaluation procedure and are now considered acceptable by the majority of CPMP Members. (However, see section 5, overall conclusions on Quality.) Both pH limit and the limit for headspace oxygen (NMT 0.5%) are supported by batch analyses and stability data. The associated test methods in these specifications have been adequately validated and are acceptable for a product of this type. Batch analyses data are still required to be submitted for the first two commercial batches released in Europe.

For the reconstituted solution – The tests include determination of the radiochemical impurities (by HPLC) for which the limits proposed have been justified. Peptide purity/impurities are also determined in the reconstituted and boiled kit. The limits proposed have been supported by batch analyses data and reference to toxicological studies.

Stability of the Product

Stability studies have been conducted on the product (kit) at ≤-10°C with an accelerated storage condition at 5°C ± 3°C. Storage at ≤-10°C is required as a result of radiochemical purity not meeting specification at the elevated temperature. Stress studies involved exposure of the product to extremes of light and heat and these conclude that the kit is sensitive to high temperatures.

Degradation products which were typically present in technetium ^{99m}Tc-depreotide solution are ^{99m}Tc "non-mobiles", which are degradation products which are not mobile in a chromatographic sense, that is, they remain at the baseline. These non-mobiles were present initially at concentrations in the region of 4-6%, although they reached 6-8% after 5 hours storage at room temperature. In addition ^{99m}Tc hydrophilic impurities (^{99m}Tc-pertechnetate, glucoheptonate and edetate) were present at 3% but there was no apparent trend for these to increase. Monitoring of the impurities species 'k', 'z' and other greatest single unknown impurity did not commence until 20-23 months into the stability programme. Four time-points were monitored either on a monthly or two monthly basis to provide information. All met the proposed specification.

Results for P1066 were well within specification at 24 months. Based on the batch data presented consideration should be given to tightening the specification for P1066, species 'k' and 'z' when more experience is gained. It should be pointed out that the main degradant P1066 is not expected to form a complex with ^{99m}Tc. Technetium ^{99m}Tc depreotide exists as two isomers - '*syn*' and '*anti*', and during storage the '*syn*' isomer increases slightly. Both isomers have a high binding affinity with the somatostatin receptors. The HPLC method is used to determine both isomers.

A shelf life of 24 months, when the finished product is stored at ≤-10°C, was proposed. Numerous changes took place during the stability programme which included upgrades to analytical methods, change of radiometric detector, change to reconstitution/incubation process for the reconstituted product, change in primary storage condition for the stability protocol, etc. Additional data were generated (during the procedure) from approximately 24 months into the stability programme for pilot scale batches to cover these changes. The proposed shelf life of 24 months is not yet supported, but an 18 month shelf-life could be granted on the basis of the data provided to date, although a follow-up measure is agreed that further data is still required from the first three commercial batches released in Europe.

As the product is to be stored at ≤ ⁻10°C and shipped at ≤+8°C a freeze-thaw cycling stability study was performed (≤⁻10°C to +5 ±3°C) and the results demonstrate that the product can withstand several freeze-thaw cycles.

Stability of the reconstituted product

In-use studies were carried out at ambient temperature using both TLC and HPLC methods to determine the purity. The finished product is allowed to equilibrate to room temperature prior to reconstitution and after reconstitution the product is stored "at ambient room temperature". As this is not in line with the ICH guideline on stability testing of new active substances and medicinal products, a new study at 25°C was initiated and the results provided demonstrate that the reconstituted product is stable for 5 hours at 25°C. An additional study clearly demonstrates that the reconstituted product should not be refrigerated prior to use. The SPC and labels include instructions to store the reconstituted solution for injection for no more than 5 hours at 15 – 25°C, but storage in a refrigerator is not specifically contra-indicated.

3. Toxico-pharmacological aspects

Pharmacodynamics

Natural somatostatin (SST) is a cyclic tetradecapeptide produced by the hypothalamus and pancreas. SST is known to modulate growth hormone release and inhibit the release of thyrotropin. Additional physiologic activities include inhibition of the release of glucagon, insulin and gastrin. In the GIT, SST decreases intestinal absorption, blood flow and mucosal cell proliferation. Receptors for SST have been found in the CNS, pituitary, pancreas, and in the mucosa of the GIT. Five subtypes of human SST receptors (SSTR) have been cloned, SSTR 1-5. Some tumours and their metastases express SSTR to a greater extent than normal tissue, these include: small-cell lung cancer (SCLC), non-small cell lung cancer (NSCLC), endocrine pancreatic tumours, metastatic carcinoids, growth hormone-producing pituitary adenomas, paragangliomas, lymphomas (mainly Hodgkin's), astrocytomas and meningeomas as well as part of the colorectal, breast and prostate cancers.

The depreotide trifluoroacetate in the Kit for the Preparation of Technetium (^{99m}Tc) depreotide is a synthetic, linear tetrapeptide attached to one of the residues of a cyclic hexapeptide, which in turn contains an amino acid sequence similar to the presumed bioactive sequence of native SST. Upon reconstitution with Sodium Pertechnetate Tc ^{99m} Injection and heating, a co-ordination complex forms with amino acid residues in the linear tetrapeptide. The Technetium (^{99m}Tc) depreotide will bind to the SSTR expressed with high density on certain tumour types, and the γ-radiation emitted (with short $t_{½}$) will aid in the scintigraphic imaging of these tumours. The proposed clinical intravenous dose of ^{99m}Tc-depreotide is 47 µg (originally nominally 50 µg) peptide. Since the product is a diagnostic imaging agent, a single administration is envisaged.

Pharmacodynamic effects relating to proposed indications:

A number of studies have been submitted which illustrate the binding of depreotide, and its *syn* and *anti* isomers (present in the ratio *syn/anti* = 0.074) to SSTR and the use of ^{99m}Tc-depreotide in binding and imaging studies.

Binding affinity studies were carried out against a variety of human tumour cell lines, tumour explants and cloned receptor sub-types. Findings suggest that ^{99m}Tc-depreotide has the highest affinity for human SCLC (85% saturation of binding sites) and NSCLC, followed by the lymphomas (50% saturation of binding sites). Inhibition/correlation of binding by ^{125}I-SST and ^{111}In-octreotide indicate the specificity of ^{99m}Tc-depreotide to SSTR. In the cell lines expressing specific receptor subtypes, it was shown that ^{99m}Tc-depreotide is specific for SSTR subtypes 2, 3, 5 and not SSTR subtypes 1 and 4. In the human SCLC cell line NCI-H69 expressing SSTR at high density (2482 fmol/mg membrane protein) depreotide inhibited ^{125}I-SST binding with IC_{50} = 6.1 nM. Overall ^{99m}Tc-depreotide had high *in vitro* binding capacity to human breast cancer, small cell lung tumours and lymphoma but lower binding capacity to non-small cell lung cancer, colon and pancreatic cancer.

The uptake of ^{99m}Tc-depreotide by rat pancreatic and human SCLC tumour xenografts was measured in mice and rats respectively, following doses of 1.0 µg peptide/kg iv, the *syn* and *anti* isomers and different radiochemical purities of the peptide were also tested, as was the inhibitory potential of octreotide. The results of these studies demonstrate that slight changes in radiochemical purity have little impact on SSTR affinity, the *anti* isomer has higher affinity than the *syn* (being approximately 6 times more potent) and that excess octreotide inhibits this binding (74%) of both the *syn* and *anti* isomers which indicates that both isomers have a high degree of specificity for interaction with somatostatin receptors. The results also

support a receptor-peptide interaction as the mechanism by which ^{99m}Tc-depreotide may target receptor-expressing carcinomas of the lung clinically. In addition, the tumour/blood and tumour/muscle ratios and the gamma scintigraphy pictures suggest a potential for good differentiation of tumour in clinical trials.

The pharmacodynamic testing programme has demonstrated the high affinity binding of ^{99m}Tc-depreotide to SSTR, *in vitro* and *in vivo* and the potential of this product in the proposed indication. Many of the *in vitro* studies use one point estimates to calculate Bmax and Kd due mainly to insufficient tissue being available. Whilst this is not an ideal situation, it is accepted that a more than accurate estimation would not have been possible in the experimental settings outlined.

General pharmacodynamics:

The binding of technetium ^{99m}Tc-depreotide to SSTR in human microvascular endothelial cell (HMVEC) membranes from both lung and dermal tissue was estimated by single point analysis and was similar to that seen with lymphoma cell lines.

A direct comparison of the effect of depreotide and SST on arginine-induced glucagon release in rat was performed. Depreotide at a dose of 1 μg/kg (1x maximal human dose, MHD), or 3 μg/kg (3x MHD), does not inhibit the arginine-induced glucagon release. However, depreotide doses of 15 x MHD suppress glucagon release, on average, by 24%. In order to obtain the same pharmacological effect as SST, depreotide had to be administered at 50 x MHD. Thus, depreotide is clearly less potent than SST.

The effects of the product on the CNS, cardiac and respiratory system were not investigated in safety pharmacology studies.

Pharmacokinetics

Biodistribution, metabolism and excretion of ^{99m}Tc-depreotide administered intravenously were investigated in rat, rabbit and monkey. The kinetics and biodistribution of unlabeled (i.e. [^{3}H]-labelled) peptide were also investigated since more than 85% of the clinical dose is unlabelled peptide.

Biodistribution:

Analyses capable of resolving the *syn* and *anti* isomeric forms of ^{99m}Tc-depreotide were conducted using RP-HPLC/ITLC-SG methods.

The studies in the rat suggest that depreotide is rapidly cleared from the plasma and that exposures are similar in males and females. The $T_{½}$ of the ^{3}H-depreotide is nearly four times and the $T_{½}$ nearly twice that of ^{99m}Tc-depreotide (6.8 vs. 1.8 min and 58 vs. 34 min respectively). This is thought to be due to the greater number of ionisable groups on the ^{3}H-depreotide. This is not considered significant since ^{3}H-depreotide was not used in safety studies. Studies were carried out at two dose levels (1 & 20 μg/kg) and essentially equivalent values for these key pharmacokinetic parameters are obtained using either dose. Consistent with this, the blood elimination curves for the 1 μg/kg and 20 μg/kg dose levels in male rats are super-imposable. Based on these studies, it is concluded that the pharmacokinetic properties of ^{99m}Tc-depreotide are not dose-dependent.

A further study was carried out in rats with experimental renal dysfunction (ERD), i.e. renal ligation. Exposure to ^{99m}Tc-depreotide was greatly increased, mainly due to a 5-fold reduction in clearance and a subsequent high uptake into the liver and skeleton. This would suggest that elimination is primarily renal and that no secondary or compensatory excretion pathway is available.

In two additional studies six NZW rabbits (3m, 3f) and one female rhesus monkey received 1.0 □g/kg iv, the pharmacokinetic parameters suggest similar handling of ^{99m}Tc-depreotide to the rat model, i.e. rapid distribution and clearance.

Biodistribution of ^{99m}Tc-depreotide was monitored in the above studies by tissue sampling and by gamma scintigraphy. In rats, distribution is mainly to the kidneys and urine, except in ERD where the liver is the main target organ, distribution is largely unaffected by labelling except from a slight increase in liver exposure. ^{99m}Tc-depreotide was also detected in the spinal and long bones as well as the bone marrow, but was not quantified. Urinary excretion is estimated at 27%, faecal at 21%. The rabbit and monkey showed similar distribution patterns to the rat.

^{99m}Tc-depreotide was incubated with human (healthy volunteer) whole blood which was subsequently fractionated. It was found that the bulk of the ^{99m}Tc-depreotide remained in the plasma (97.27%), while the rest (1.25%) was associated with blood cells, platelets etc.

Metabolism:

After ^{99m}Tc-depreotide was incubated in rat and human plasma, approximately 70% total peptide was recovered, <2% in either species was parent peptide. The remainder was seen as '"oxidative"' products on RP-HPLC, but no further identification was carried out.

In a study in rats (1.0-20 µg/kg iv) no metabolites were detected in the bile. In the plasma several more lipophilic structurally related metabolites were identified by RP-HPLC. Approximately 20% of the dose is eliminated in the urine in four hours, and this includes several more hydrophilic species identified by RP-HPLC. The main metabolite seen on incubation of ^{99m}Tc-depreotide in rat plasma *in vitro* was not detected in the plasma or urine of humans or in the plasma, bile or urine of rats. This metabolite was produced by incubation of ^{99m}Tc-depreotide with rat kidney homogenate, but not with kidney slices or with kidney homogenate treated with aprotinin. Since metabolism following *in vivo* administration accounts for <1% of the injected dose, the significance of the observed metabolism after prolonged incubation with rat plasma or with kidney homogenates is debatable.

Toxicology

The scientific literature provides ample evidence for the role of SST as a physiologic mediator (and hence for the presence of SSTR) in the rat, rabbit and man. Studies also show that SST and SSTR are involved in similar biochemical and biological processes in these species, and that the data provided support the use of rats and rabbits as appropriate animal models to assess the toxic potential of ^{99m}Tc-depreotide.

Single dose toxicity

In Swiss albino mice body weight was not affected up to 14 days after administration of 0-300-1000 µg/kg iv ^{99m}Tc-depreotide. Some lethargy & piloerection were seen immediately post-dose. Although splenic atrophy was observed in males this was without gross or histological correlates. Low group sizes could be the reason for the missing significant difference in spleen weight. No significant changes were seen in haematology, clinical chemistry or organ weights, and there were no macroscopic or microscopic findings. Therefore the no-observable-effect-level (NOEL) was 1000 µg/kg iv.

In NZW rabbits administered 0-200-600 µg/kg iv, some statistically significant fluctuations in clinical chemistry parameters were noted, but these were not considered biologically significant (most notably a reduced CPK, LDH and BUN). Haematology, organ weights and macroscopic and microscopic observations were normal in all groups. The NOEL was 600 µg/kg iv.

These studies (conducted according to GLP) suggest that there is no acute toxicological hazard to man from the use of ^{99m}Tc-depreotide in the proposed indication.

Repeated dose toxicity

A series of repeated dose toxicity studies were carried out in the rat, in all cases the animals were given ^{99m}Tc-depreotide reconstituted in decayed generator eluate or a suitable control. In two studies, rats received doses of 0-30-100 µg/kg/day iv for 10 days and 14 days respectively. In a third study rats received doses of 0-40-100 µg/kg/day iv for 14 days. Results were similar for all three studies. There were no drug-related deaths or changes in body weight gain. Clinical observations were normal apart from occasional injection site discoloration/haematoma. Some fluctuations in haematology and clinical chemistry parameters were seen, most notably: an apparent, slight, dose related increase in RBC, Hb and Hct in females; a slight reduction in LDH in treated males; a slight reduction in CPK and increase in AP in treated females. A reduction in the albumin/globulin ratio and in total cholesterol, associated with a fall in relative liver weight was seen in top dose females in the first study only, this is thought to be drug related but was not repeatable and is therefore probably not biologically significant. Apart from this liver weight change, all necropsy findings were within normal parameters.

In three studies in rabbits, the animals (6/sex/group) received 0-100-400 µg/kg/day iv for 10 or 14 days and 0-30-100 µg/kg/day iv for 14 days. Results were similar for all three studies. There were no drug-related deaths or changes in body weight gain. Clinical observations were normal apart from occasional injection site discoloration/haematoma. Some fluctuations in haematology and clinical chemistry parameters were seen, most notably: an apparent, slight, an increase in RBC, Hb and Hct in top dose females; a slight reduction in LDH in treated males; a slight reduction in CPK, SGOT and SGPT in treated females. A reduction in the albumin/globulin ratio, associated with a fall in relative liver weight was seen in top dose females in the second study, a lesser fall in liver weight was seen in the third study which was not associated with reduced albumin, this is thought to be drug-related but not biologically significant. Apart from this liver weight change, all necropsy findings were within normal parameters.

In both species small changes in RBC, Hb, Hct, CPK and albumin in females and reduced LDH in males, occurred only at 100X MHD and are therefore not considered as clinically relevant. The NOEL (no-observable-effect-level) in all the studies was at least 100 times the anticipated human dose of 1 µg/kg.

Genotoxicity

Depreotide and ^{99m}Tc-depreotide were not mutagenic in the bacterial reverse mutation assay (*S. typhimurium* and *E. coli* WP2uvrA) or in the mammalian cell forward mutation assay (mouse lymphoma L5178Y TK+/- cell line) in the presence or absence of metabolic activation.

Furthermore, ^{99m}Tc-depreotide did not induce any genotoxic effects *in vivo* in the bone marrow micronucleus test at high doses (4 mg/kg iv)**.**

Carcinogenicity

No data were submitted concerning the carcinogenic potential of ^{99m}Tc-depreotide. This is considered acceptable because the product is intended for single use in the clinical setting, is not mutagenic or clastogenic and there are no structural alerts.

Reproduction toxicity

No studies were submitted concerning the potential reproductive toxicity of ^{99m}Tc-depreotide. However, as the administration of a radionuclide to a pregnant woman would result in exposure of the foetus to a radiation dose, a contraindication has been included in section 4.3 of the SPC for pregnancy and lactation. Section 4.6 of the SPC also emphasises this contraindication and that, as it is not known whether ^{99m}Tc-depreotide is excreted in human milk, administration is contraindicated during lactation.

Local tolerance and immunotoxicity

A perivascular irritation study was carried out in NZW rabbits. There was no dermal erythema, oedema, vascular congestion, epithelial erosion, or local leukocytic infiltration. On a scale of 1-4, ^{99m}Tc-depreotide was assigned an irritation index of +0.3 with respect to vehicle and is thus considered as non-irritant.

^{99m}Tc-depreotide was non-antigenic in a standard guinea pig systemic antigenicity model.

The compatibility of ^{99m}Tc-depreotide with human blood was tested *in vitro*. Anti-coagulated human blood or human serum was mixed with ^{99m}Tc-depreotide to a final concentration of 2-4-fold anticipated clinical levels. There was no difference seen between vehicle and peptide, all measured parameters (Hb, Hct, RBC, WBC, platelets, sedimentation rate, total protein) were within normal ranges suggesting that the product is compatible with human blood.

The local tolerability of the product, ^{99m}Tc-depreotide, is considered adequate for the proposed indication.

Ecotoxicity

The environmental effects of potential release are predicted to be minimal and of no concern.

Discussion on toxico-pharmacological aspects

It was shown in both *in vivo* and *in vitro* studies that ^{99m}Tc-depreotide and both its isomers have high affinity to various tumours and tumour cell lines expressing SSTR. It was also shown that depreotide had higher affinity for SSTR subtypes 2, 3 and 5. The high binding affinity of ^{99m}Tc-depreotide to the SSTR 3 may be clinically relevant since this subtype 3 appears to be over-expressed in many tumours. Furthermore the spectrum of tumours targeted by this radio-pharmaceutical is not affected by isomer conversion *in vivo*. The data demonstrate that 85% and 94% radiochemical purity ^{99m}Tc-depreotide were equally effective in terms of somatostatin receptor affinity.

Although no safety pharmacology studies were carried out the overall data do not give rise for concerns.

No drug interaction studies have been performed, which is acceptable, considering the proposed indication.

Although there were species dependent difference in some pharmacokinetic parameters, ^{99m}Tc-depreotide was rapidly cleared from the blood pool ($t_{1/2a}$ from 1.6 to 2.4 min) and nearly completely from blood (>98%) within 4 hours in all three species investigated. Furthermore, the volume of the central compartment exceeded the estimated blood volume, and the volume of distribution at steady state approximated a volume in excess of the extra cellular fluid volume, indicating widespread distribution (particularly to the kidneys, liver and GIT). The majority of the radioactivity is eliminated via the urine.

Less than 2% of ^{99m}Tc-depreotide binds to blood cells and plasma proteins and no alterations in haematological parameters were noted.

There was evidence of the formation of more lipophilic and more hydrophilic species, but metabolites were not further characterised.

The compound accumulated in renally impaired animals. Although the human data indicate that there is no need to reduce the dose in renally impaired patients (see Part IV.A), the following warning has been included in the SPC, under Section 4.4 Special warnings and special precautions for use "Care should be exercised in patients with impaired renal function, due to lower renal excretion and probable increase in exposure to radioactivity."

In single and repeat (up to 14 days) dose toxicity studies, clinically relevant toxic effects were not observed doses up to 100 times the anticipated human dose of 1 µg/kg.

No studies were submitted concerning the potential reproductive toxicity of ^{99m}Tc-depreotide. Since, the use of the product results in administration of a radionuclide, contraindications for both pregnancy and lactation has been included in the SPC.

^{99m}Tc-depreotide was not genotoxic *in vitro* or *in vivo*.

No data were submitted concerning the carcinogenic potential of ^{99m}Tc-depreotide. This is considered acceptable because the product is intended for single use.

^{99m}Tc-depreotide was non-irritant in rabbits and non-antigenic in guinea pigs.

The environmental effects of potential release are predicted to be minimal and of no concern.

The species used in studies for Part III of the dossier represent laboratory animals of known quality and homogeneity, commonly used in pharmacological/toxicological research, which were shown to be relevant to the product and indication. All pivotal safety studies were conducted according to GLP with the exception of 10-day repeated dose toxicity studies in rats and rabbits. The scientific standards and the performance of these studies are sufficient to assess the toxicological effects of ^{99m}Tc-depreotide in laboratory animals and suggest a good safety profile for the drug's use in man.

4. Clinical aspects

Clinical pharmacology

The Applicant conducted 5 studies in healthy volunteers and in patients with renal or hepatic impairment:

Study number	Type of study	Doses peptide/ ^{99m}Tc-depreotide intravenous injection	Subjects
Study 829-13	Phase I Pharmacodynamic	50 µg peptide / 0 MBq	9 healthy volunteers
Study 829-10	Phase I Pharmacokinetic	35-50 µg peptide / 307-400 MBq	17 healthy volunteers
Study 829-12	Phase I Pharmacokinetic	50 µg peptide / 303-566 MBq	23, of these 12 healthy volunteers and 11 patients
Studies: 829-23, 829-22 & 829-30Iia	Phase II Optimal dose Radiation dosimetry	10, 20 or 50 µg peptide / 185, 370 or 740 MBq	46 patients

Pharmacodynamics

Study 829-13 evaluated the pharmacodynamic effects of unlabelled depreotide and octreotide on the glucose tolerance response in 9 normal volunteers receiving 50 µg of each peptide intravenously and after no injection, followed by an oral glucose tolerance test. These results suggest that depreotide, does not alter the physiological response to a glucose challenge.

Pharmacokinetics

Study 829-10 evaluated the biodistribution, uptake and clearance half-times in healthy volunteers Seventeen healthy volunteers received approximately 50 µg depreotide labelled with approximately 370 MBq ^{99m}Tc. Two subjects could not be evaluated for clearance or dosimetry due to incomplete data collection and three subjects were not included in the analyses due to suspected errors in the analyses of the samples. Most of the results are calculated from data of 12 subjects. Serial blood samples were collected at multiple time points post-injection up to 24 hours, and urine was collected over 24 hours. Whole body scintigraphy at 10 minutes, 1, 2, 4 and 18-24 hours post-injection was performed in order to assess whole body biodistribution of ^{99m}Tc-depreotide.

Blood activity levels following intravenous injection exhibited a triphasic decay with a rapid decline in radioactivity with a half-life of 3.7 minutes (median), followed by a more gradual decline with a half-life of 35.2 minutes (median), and a slow decline with a half-life of 17.9 hours (median). Urinary excretion accounted for 5.3 - 7.7% of the total injected dose (ID), and the average renal clearance of radioactivity was 0.255 ml/min/kg. The total clearance was calculated to be 3.89 ml/min/kg.

The body weight-normalised volume of distribution at steady state and volume of distribution of the central compartment were 3.12 l/kg and 0.154 l/kg, respectively. Serial scintigraphic body images indicated the highest activities (% ID) in the kidneys (13%), liver (10%), pelvic area (6.3%), and lungs (6.12%) at 10 minutes post-injection, and during the first 24 hours relative activity in these regions remained nearly constant.

Study 829-12 aimed to evaluate the pharmacokinetics and radiation dosimetry of ^{99m}Tc-depreotide. Of 23 subjects 12 healthy volunteers (6 men and 6 women), 4 patients with renal impairment (2 men and 2 women), 5 patients with hepatic impairment (4 men and 1 women) and 2 patients having lung cancer (both men) received 50 µg peptide labelled with 555-740 MBq ^{99m}Tc intravenously. Blood and urine samples were collected and scintigraphic body images were obtained during the first 24 hours. Metabolites in plasma and urine were determined by gamma-counting of the RP-HPLC separated fractions. Plasma protein binding of ^{99m}Tc-depreotide was determined by gel exclusion column chromatography. Antibodies at baseline and three week post-injection plasma samples was determined by an enzyme linked immunosorbent assay (ELISA) sensitive for IgG and IgM.

Plasma radioactivity indicated triphasic decay, with mean half-life values of 4.4 minutes, 48.7 minutes and 19.8 hours. Systemic clearance was 2.01 ml/minute/kg and renal clearance was 0.34 ml/minute/kg. This relatively low renal clearance indicated extra-renal elimination. The volume of distribution was large (1.5 l/kg) and exceeded total body water volume. Urinary excretion accounted for 1-18% of the injected dose.

Plasma protein binding and HPLC analyses of plasma and urine samples indicated that >90% of the radioactivity eluted from the columns in fractions corresponding to the parent drug, thus little circulating plasma radioactivity represents metabolites. The majority of radioactivity in urine also eluted in HPLC fractions corresponded to those of the parent drug. Mean plasma protein binding was 12.1% and the patients were comparable to healthy volunteers.

In these studies ^{99m}Tc-depreotide did not cause any generation of human IgG or IgM depreotide-specific antibodies.

Further discussion of the impurities in the radioactive solution and their distribution to non-target organs was requested during the evaluation procedure. The impurities were not intentionally included in the technetium (^{99m}Tc) depreotide administered in the human studies. However, the impurities are typically present in all preparations of technetium (^{99m}Tc) depreotide based on release and stability testing performed by the applicant, suggesting that the clinical preparations would have contained a typical impurity profile for technetium (^{99m}Tc) depreotide. Thus, while not specifically monitored, it is likely that technetium (^{99m}Tc) depreotide administered to clinical subjects would contain representative impurities and the biodistribution of those impurities would have been included in the dosimetry calculations.

The sponsor has compared biodistribution of technetium (^{99m}Tc) depreotide (in the AR42J tumor xenograft nude mouse model) of 80% radiochemical purity by HPLC (20% impurities) and 95% radiochemical purity by HPLC (5% impurities). Both preparations met the NLT 90% radiochemical purity limit as determined by TLC. No significant change in biological performance and biodistribution was noted between the 80% and 95% radiochemical purity by HPLC preparations. Also, *in vitro* studies demonstrated that both 80% and 95% radiochemical purity by HPLC preparations have equal affinity for somatostatin receptors by AR42J tumour membrane preparations. Thus, the presence of 20% impurities in the technetium (^{99m}Tc) depreotide had negligible impact on binding affinity and biodistribution in animal and *in vitro* studies.

In summary, the impurities have been present in preparations used for clinical studies for calculation of human biodistribution of technetium (^{99m}Tc) depreotide.

Dose response studies

Optimal dose, radiation dosimetry and imaging time

The aim of **study 829-23** was to assess the optimal dose ranges for the peptide and ^{99m}Tc. Three different doses of the peptide, 10, 20 or 50 µg were labelled with three different levels of radioactivity - either 185, 370 or 740 MBq (5, 10 or 20 mCi). The study was designed as a multi-centre, randomised, and parallel group 3 X 3 factorial design in 45 patients with clinically documented or suspected somatostatin receptor-expressing tumours. ^{99m}Tc-depreotide images were evaluated relative to OctresScan® images. Similar results were obtained with all doses with no safety advantages of the lower doses. The 50 µg peptide dose labelled with 740 MBq (20 mCi) was selected as the optimal dose because 50 µg provides a larger capacity to carry the radiolabel as it decays than lower doses, and 50 µg is the minimal dose that can be used to make a rugged kit practical for clinical use. The 740 MBq (20 mCi) ^{99m}Tc dose was chosen because it falls within the lower end of the standard ^{99m}Tc dose range which is 555-1110 MBq (15-30 mCi), was a safe dose in terms of radiation dosimetry and produces a photon flux necessary for Spect imaging. With a lower dose patients would have to be scanned for longer periods and difficulties in lying still may cause "blurred" scintigraphic images.

Radiation absorbed doses were calculated on the basis of blood activities, urine activities and gamma camera data from study 829-12 after injection of ^{99m}Tc-depreotide to 22 subjects (11 subjects and 11 patients (5 with liver disease, 4 with renal disease and 2 with lung cancer). The MIRD method of the Society of Nuclear Medicine was used for absorbed dose calculations.

Only small differences, which were not significant, were seen between the estimated organ doses for normal and diseased patients.

The biodistribution of total activity at 10 minutes post-injection showed the highest values for the abdomen (59%). The highest values were for the liver (15.3%) and kidneys (7.4% in left and 8.2% in right), and the lowest for the thyroid (0.4-0.8%). A range of 3.5-5.8% was associated with the lungs. During the 24 hours post-injection period, activity remained nearly constant in these regions. Tissue

distribution of ^{99m}Tc-depreotide in renally or hepatically impaired patients or in patients with lung cancer was similar to the tissue distribution seen in healthy subjects although the numbers of patients were limited. The cumulative urinary excretion was in the range of 4.2% to 22.7% with an average value for urinary excretion of 10.4%. The mean overall elimination half-life was 5.1hr (0.5-15.6hr).

The highest organ doses were calculated for the kidneys (0.090 mGy/MBq), the spleen (0.042 mGy/MBq) and the testes (0.031 mGy/MBq). The dose estimates for all other organs were lower than 0.03 mGy/MBq. The effective dose equivalent was 0.023 mSv/MBq.

The average maximum uptake in the testes was estimated to be 1.3% ID. Activity was taken up and retained in the marrow in all subjects with a substantial bone marrow residence time (2.82 hours), but modest dose of 0.021 mGy/MBq with large inter-individual variation.

During the evaluation procedure, comparative data for conventional CT, spiral CT and NeoSpect were presented. The results are shown in Table 1. The proposed indication for NeoSpect is scintigraphic imaging of suspected malignant SPNs in combination with CT scan or chest X-ray. The radiation dose from NeoSpect alone does not appear to pose a safety risk for radiation-induced injury to tissues and following the addition of the radiation dose from a CT scan, the risk still remains within the accepted limits. Using spiral CT instead of conventional CT may be an alternative approach to keeping the radiation dose to a minimum.

Table 1. Comparison of Radiation Doses to Tissues from NeoSpect, Chest CT, and Spiral CT Scans

Tissue	Estimated Absorbed Radiation Dose from 740 MBq NeoSpect (mGy)	Estimated Absorbed Radiation Dose from Chest CT (mGy)	Estimated Absorbed Radiation Dose from Spiral CT (mGy)[4]
Lungs	10	19.2 - 24.4	10 – 16
Bone Marrow	16	3.95 - 5.79	3.3 – 4.7
Breast	1[3]	23.5 - 27.1	9.7 – 14
Thyroid	18	1.93 – 3.06	2.3 – 5.2[5]

[3]The estimated absorbed radiation dose for breast is ~0.001 mGy/MBq (data for breast not included in Table 2).

[4]Data extracted from Expert statement, attachment 4 "Organ doses in spiral CT of the chest**"**.

[5]Dependent on the actual slice location in the lower part of the collum area.

In **study 829-22** the imaging time was of no consequence for image quality in 13/26 patients with lung cancer while images obtained between 30 mins – 2 hours after injection were optimal in all the other 13 patients. Most images considered optimal by the investigator were obtained between 3 and 5 hours after injection in study 829-30IIa.

In conclusion, studies in healthy volunteers and in patients demonstrated that technetium (^{99m}Tc) depreotide confers 3-compartment pharmacokinetics with a distribution half-life of less than 5 minutes, a terminal half-life of about 20 hours and a steady state volume of distribution of 1.5-3 L/kg. Total clearance averaged 2-4 ml/min/kg while renal clearance averaged about 0.3ml/min/kg and thus accounts for only a small proportion of the rapid blood clearance. External whole-body gamma scintigraphy showed highest localisation of radioactivity in the abdomen (in the liver and kidneys) but this did not appear to affect the imaging capacity of technetium (^{99m}Tc) depreotide for the lungs. The visualisation of lung cancer lesions by scintigraphy is based on higher tumour uptake relative to normal lung tissue i.e., background. This would result in a higher radiation dose to the normal lung in patients however the absorbed dose calculation (0.014 mSv/MBq) does not suggest any burden.

The overall mean elimination half-life for Technetium (^{99m}Tc) depreotide calculated from dosimetric data was 5.1 hours.

A statistically significant difference for total clearance between men and women was seen in study 829-12, although as this resulted from one high value it is not considered to be clinically relevant.

Tissue distribution of technetium (^{99m}Tc) depreotide in patients with impaired renal or hepatic function, or in lung cancer patients, was not appreciably different from that observed in healthy subjects although the smaller numbers of patients in the specific groups precluded adequate statistical evaluation.

Typically >90% of the plasma radioactivity corresponds to the parent drug with absence of metabolites. Plasma protein binding of technetium (^{99m}Tc) depreotide was about 12%.

Using ELISA sensitive for IgG and IgM, no antibodies were detected (single injections in 18 volunteers and more than one injection in 13 of these).

The clinical pharmacology of ^{99m}Tc-depreotide has been adequately studied. Appropriate special precautions have been included in the SPC for renal and hepatic impairment.

Clinical efficacy

The clinical trials were performed according to GCP standards.

Main studies

Study number	**Type of study**	**Doses peptide/ ^{99m}Tc depreotide intravenous inj.**	**No. of patients**	**Study design**		
				reference method	**comparator**	**evaluation of ^{99m}Tc depreotide images**
Study 829-30IIa	non-controlled, phase II, O, SC, 1 dose safety	50µg / 592-814 MBq	13, 12 patients with NSCLC were evaluated for efficacy	histopatho-logy	FDG-PET	investigator read
Study 829-22	non-controlled, phase II, MC, 1 dose, unblinded, non-R, safety	30-50µg / 240-851 MBq	141, 111 patients were evaluated for efficacy	all confirmato ry diagnostic procedures	other different confirmatory diagnostic procedures	investigator read

Study number	Type of study	Doses peptide/ ^{99m}Tc depreotide intravenous inj.	No. of patients	Study design		
				reference method	comparator	evaluation of ^{99m}Tc depreotide images
Study 829-34A	non-controlled, phase III, MC, 1 dose, comparative trial, safety	18.3-50.0µg / 548-1103 MBq	128, 112 patients were evaluated for efficacy	histopatho-logy	CT 3 blinded readers and investigator read	3 blinded readers and investigator read
Study 829-34B	non-controlled, phase III, MC, 1 dose, comparative trial, safety	20.0-50.0µg / 448-1214 MBq	142, 114 patients were evaluated for efficacy	histopatho-logy	CT 3 blinded readers and investigator read	3 blinded readers and investigator read
Study 829-P	non-controlled, phase III, MC, 1 dose, comparative trial	18.3-50.0µg / 448-1214 MBq	127 patients from the two above mentioned studies	histopatho-logy	CT 3 blinded readers and investigator read conducted in the two above mentioned studies	3 blinded readers in presence of CT or chest X-ray
Study P829-30A	non-controlled phase III, MC, 1 dose,	7-50 µg / 455-870 MBq	117, 115 patients were evaluated for efficacy	FIC diagnosis	^{111}In pentre-otide investigator read	investigator read 3 blinded readers
Study P829-30B	non-controlled, phase III, MC, 1 dose,	12.5-50 µg / 403-995 MBq	135, 128 patients were evaluated for efficacy	FIC diagnosis	^{111}In pentre-otide investigator read	investigator read 3 blinded readers
Study 829-40	Non-controlled, phase III, MC			Histopatho logy		3 blinded readers in presence of CT or chest X-ray

MC: multi centre; SC: Single centre; O; open label; R: randomised; FIC diagnosis: final institutional clinical diagnosis;

- The total number of patients included in the studies: 712 + 59 patients
- Patients evaluated for efficacy: 637 +49 patients
- Patients with known or suspected cancer of the lung: 348 patients

The Phase III **studies 829-34A and 829-34B** were identical with respect to study objectives and enrolled together 258 patients with suspected lung cancer. Patients diagnosed with the clinically relevant criteria of solitary pulmonary nodules (SPN) were included in the analysis presented in the study 829-P.

Both studies used a within-patient comparative design to evaluate the efficacy of ^{99m}Tc-depreotide scintigraphy for the detection/localisation of the main presenting lesion in patients with a suspicion of lung cancer. The patients underwent planar imaging in multiple views as well as SPECT and whole body imaging. Images were assessed by the investigator as well as by three independent nuclear medicine practitioners, who were blinded to all other clinical information. All patients had CT imaging performed, also assessed by three independent radiologists who were blinded to all other clinical information. The scintigraphic images were evaluated as being positive or negative for the regions right/left upper and lower, right middle, right/left hilar and mediastinum. Efficacy was evaluated by calculation of the sensitivity, specificity and agreement of ^{99m}Tc-depreotide scintigraphy, relative to histopathological diagnosis of the main presenting lung lesion. The investigator using the histopathological results determined lesion location. The sensitivity and specificity of ^{99m}Tc-depreotide was compared to CT imaging, relative to histopathological diagnosis.

The efficacy population comprised all patients who were enrolled and who received the ^{99m}Tc-depreotide and successfully completed both scintigraphy and CT imaging, and for whom there was an available histopathological diagnosis for the main presenting region - in total 212 patients.

The normal approximation to the binomial was used to test the hypothesis:

$H_0 : \pi_T \leq 0.7$ versus the one-sided alternative $H_1 : \pi_T > 0.7$ where π_T is the proportion of patients with agreement between technetium (^{99m} Tc) depreotide results and histopathology results. Overall p-values were calculated for the majority blind read and the investigator read result. The value of 0.7 (rather than the initial value of 0.8) was chosen because agreement rates between histopathology and CT are said to be no better than 65%.

Because performance of CT was a prerequisite for histopathology the comparison between technetium (^{99m} Tc) depreotide and CT was kept as a secondary efficacy endpoint. The sensitivity and specificity of CT relative to histopathology were calculated for the main presenting lesion.

In order to take prevalence of the disease into account the negative predictive value (NPV) and the positive predictive value (PPV) of technetium (^{99m}Tc) depreotide relative to histopathology results for the main presenting lesion were calculated using positive and negative likelihood ratios (PLR & NLR) so PPV & NPV were defined as follows:

PLR = sensitivity/1-specificity

NLR = 1-sensitivity/specificity

PPV = prevalence x sensitivity/ (1-prevalence)(1-specificity) + (prevalence x sensitivity)

NPV = (1-prevalence)(specificity)/prevalence x(1-sensitivity) +(1-prevalence)(specificity)

Positive and negative predictive values were calculated for a range of prevalence of disease that might be characteristic of different populations being evaluated for the possibility of intra-thoracic malignancy. For fixed values of sensitivity and specificity PPV is an increasing function of prevalence and NPV is a decreasing function of prevalence.

Agreement between the three 'blind' readers of Technetium (^{99m} Tc) depreotide images was evaluated using the kappa statistic where a value of 0 indicates no agreement and 1 very good agreement.

McNemar's test was used to compare the sensitivity and specificity of Technetium (^{99m} Tc) depreotide with CT for the main presenting lesion.

The Wilcoxon Signed Rank test was used to analyse the change from pre-injection to each post-injection time point across all patients for vital signs and laboratory tests.

In the pivotal studies, subgroup analysis of patients presenting with SPN was performed (post-hoc).

Subgroup analysis was also performed in the pivotal studies in patients with renal or hepatic impairment and based on demographic subdivisions of age, gender and race.

Table 2. Efficacy Results for ^{99m}Tc-depreotide scintigraphy (from the Clinical Expert Report)

Read	Sensitivity	Specificity	Agreement	TP	TN	FP	FN	ALL
Blinded Majority								
Study 829-34A	82.1%	60.7%	76.8%	69	17	11	15	112
Study 829-34B	85.0%	57.1%	81.6%*	85	8	6	15	114
both combined	83.7%	59.5%	79.2%*	154	25	17	30	226
Investigator								
Study 829-34A	96.4%	53.6%	85.7%*	81	15	13	3	112
Study 829-34B	97.0%	57.1%	92.1%*	97	8	6	3	114
both combined	96.7%	54.8%	88.9%*	178	23	19	6	226

Data are presented for the main presenting lesion relative to histopathological results.

** $p<0.05$*

Using histopathological diagnosis as the gold standard, sensitivity and specificity were calculated, showing a larger number of false negatives read for the blinded read than for the investigator.

There were 17/226 false positive cases for the blinded read and 19/226 false positive cases for the investigators read. The false positive cases were attributed to granulomas (n=7), sarcoid (n=3), haematoma (n=1), inflammation (n=3), pneumonia/abscess (n=2), and fibrosis (n=1). Thirteen/17 cases, considered false positive in the blind read, were also considered false positive by the investigator read.

Agreement was defined as the percentage of correct diagnoses by ^{99m}Tc-depreotide using the histopathological diagnosis as gold standard. The agreement rate for the blind read was 79.2% and the agreement rate for the investigator read was 88.9%.

Positive predictive values (PPV) for the investigator read were between 49 and 69% and between 46 and 67% for the majority blind read for prevalence between 30 and 50%. Negative predictive values (NPV) for the investigator read was between 94 and 98%, and the majority blind read had a NPV between 79 and 90% for prevalence in the range of 30 to 50%.

Overall kappa statistics for pair-wise comparisons of blind readers (0.725) was indicative of a substantial inter-reader agreement.

Subgroup evaluation (post-hoc)

Subgroup analysis based on demographics (age > vs. ≤ 65yr, sex, race), renal and hepatic function and nature and size of the main presenting lesion were performed with the same end points.

Size of the primary lesion:

In the pivotal studies 829-34A & B, patients were subgrouped between patients with non-calcified solitary pulmonary nodulus (SPN) 1-3cm diameter and non-calcified SPN ≤ 6cm diameter. (Calcification is usually non-malignant and it is important to see how sensitivity and specificity vary with size.)

In study number 829-P the analyses included the same SPN• patients divided into 2 subgroups: patients with SPN and patients with non-calcified SPN < 3cm in diameter.

The analyses show that for the blind read the pooled subgroup for patients with SPN 1-3 cm is approximately 15% lower than that for all SPN patients. For the investigator's read the subgroup

sensitivity is very similar to that for all SPN. With regard to specificity, the ^{99m}Tc-depreotide scintigraphy was very similar for both subgroups as well as for both reads.

Table 3. Efficacy results for ^{99m}Tc-depreotide scintigraphy in patients with SPN - Combined data from studies 829-34A and 829-34B

Read	Sensitivity	Specificity	Agreement	T	TN	FP	FN	ALL
Blinded Majority Read								
all SPN patients	79.0%	66.7%	76.4%	7	18	9	21	127
SPN 1-3 cm	64.8%	68.0%	65.8%	3	17	8	19	79
SPN ≤ 6 cm	76.1%	69.2%	74.9%	6	18	8	21	114
Investigator Read								
all SPN patients	96.0%	59.3%	88.2%	9	16	11	4	127
SPN 1-3 cm	94.4%	60.0%	83.5%	5	17	10	3	79
SPN ≤ 6 cm	95.5%	61.5%	87.7%	8	16	10	4	114

Age, gender, race and disease history:

Agreement rate and sensitivity were similar in the two age subgroups, both for the blind read and investigator read technetium (^{99m} Tc) depreotide results respectively. Specificity results differed possibly because of number inequality (blind read 30 vs. 68.8%). Agreement rates, specificity and sensitivity were similar for males and females. There were no differences between Caucasian and non-Caucasian patients however there was imbalance in sample size.

While the data collected do not indicate that technetium (^{99m} Tc) depreotide would perform less effectively in patients with impaired renal or hepatic function the numbers were small.

Comparison of ^{99m}Tc-depreotide scintigraphy to CT:

Separate analysis comparing sensitivity and specificity for blind read ^{99m}Tc-depreotide and blind read CT, using histopathology as a reference method, were performed for all patients and SPN subgroups. Sensitivity for the CT scan majority blind read was 95.7% for all lesions, and >96% for all SPN subgroups, whereas the corresponding sensitivity for the ^{99m}Tc-depreotide scintigraphy was significantly lower ($p < 0.05$) for all SPN (solitary pulmonary nodules) subgroups. Specificity, however, was significantly lower for CT than for ^{99m}Tc-depreotide.

Table 4. Comparison of ^{99m}Tc-depreotide to CT (Majority Blind Read)

	Sensitivity	Specificity	TP	TN	FP	FN
Main presenting lesions (all patients)						
^{99m}Tc-depreotide	83.7%	59.5%	154	25	17	30
CT	95.7%	4.8%	176	2	40	8
SPN Subgroup						
^{99m}Tc-depreotide	79.0%	66.7%	79	18	9	21
CT	97.0%	7.4%	97	2	25	3
SPN Subgroup (1-3 cm)						
^{99m}Tc-depreotide	64.8%	68.0%	35	17	8	19
CT	96.3%	8.0%	52	2	23	2
SPN Subgroup ≤ 6 cm)						
^{99m}Tc-depreotide	76.1%	69.2%	67	18	8	21
CT	96.6%	7.7%	97	2	25	3

^{99m}Tc-depreotide images were read in the presence of CT scans or chest X-rays. CT scans were available for all patients (n=127) but chest X-rays were not available for 11 patients (n=116).

From a total of 127 patients included in the analyses, 65% (82 patients) had SPN ≤ 3 cm and 39 patients had SPN >3 ≤ 6 cm. The majority of lesions were non-calcified (90%). The prevalence of malignancy was 70% (100/127 patients) with adenocarcinoma (28%) and squamous cell carcinoma (27%) being the most commonly diagnosed tumour types.

CT scan alone has a high sensitivity but a very low specificity. ^{99m}Tc-depreotide scans in the presence of CT scan resulted in high sensitivity and a remarkable rise in specificity (from 7% to 63%). The sensitivity and specificity of ^{99m}Tc-depreotide with use of chest x-rays were 97% and 73%, respectively. The difference in specificity between CT scan alone and ^{99m}Tc-depreotide scan with CT scan or chest x-ray, resulted in an upward shifted PPV curve with fewer false positive results, when ^{99m}Tc-depreotide were included in the diagnostic work-up.

The negative predictive values for ^{99m}Tc-depreotide in the presence of CT scan or chest x-ray is high (between 90% and 98%) in the range of prevalences from 30-50%, indicating that a patient in this population, with a positive ^{99m}Tc-depreotide scan with a supportive CT scan or chest x-ray, has a high likelihood of malignancy.

Table 5. Efficacy results of blind readers: sensitivity, specificity, and agreement (relative to histopathology) (Study 829-P)

	Sens.	Spec.	Agreem.	TP	TN	FP	FN	Total
All SPN:								
CT alone	97%	7.4%	78%	97	2	25	3	127
^{99m}Tc-depreotide with CT	93%	63%	86.6%	83	17	10	7	127
^{99m}Tc-depreotide with x-ray	96.7%	73.1%	91.4%	87	19	7	3	116
Non-calcified SPN <3cm:								
CT alone	96.3%	8.0%	68.4%	52	2	23	2	79
^{99m}Tc-depreotide with CT	88.9%	64.0%	81.0%	48	16	9	6	79
^{99m}Tc-depreotide with x-ray	94.2%	75.0%	88.2%	49	18	6	3	76

The prevalence of malignancy was 79% in study 829-P. It was considered by the applicant that the only way to ethically conduct a study in a population having a lower prevalence of malignancy was to conduct the trial in a geographical location where granulomatous disease is prevalent, and this was done in Phoenix, Arizona, a region where coccidioidomycosis is endemic. 30 consecutive patients were studied (2 under the 829-22 protocol and 28 under the 829-34 protocol) with SPN referred to herein as the "Phoenix cohort…". Their results show that their population had a prevalence of malignancy of only 43.3 %. Nevertheless, in that population, technetium (^{99m}Tc) depreotide scintigraphy had a sensitivity and specificity of 92% and 88.2% respectively.

During the evaluation procedure it was considered necessary to investigate by further study in a low prevalence population whether NeoSpect (^{99m}Tc-depreotide) can reduce the need for biopsy in such a population, with particular reference to whether sensitivity and specificity is a function of tumour size distribution.

An additional clinical trial (829-40) was therefore performed in the south-western USA. In this geographical region, a prevalence of malignancy similar to that observed in the Phoenix cohort was anticipated.

In a multicenter, open-label clinical study, technetium (^{99m}Tc) depreotide scintigraphy was to be compared to histopathological results in patients presenting with an SPN of size less than 4 cm and suspicious for malignancy. The study was designed to demonstrate with 80% power that the diagnostic accuracy of technetium (^{99m}Tc) depreotide scintigraphy was greater than 70%. A total of 50 to 55 patients were to be enrolled. Each patient was to receive an intravenous injection of 555 – 740 MBq (15-20 mCi) technetium (^{99m}Tc) depreotide (approximately 50 µg depreotide peptide). The primary efficacy criterion was the accuracy of the technetium (^{99m}Tc) depreotide scintigraphy read blindly in combination with CT scan and/or chest x-ray relative to the histopathologic diagnosis (benign/malignant) for the suspicious SPN. Secondary efficacy analyses were performed to evaluate the sensitivity and specificity of technetium (^{99m}Tc) depreotide scintigraphy read blindly in combination with CT scan or chest x-ray for the identification of malignant solitary pulmonary nodules. Secondary analyses of sensitivity, specificity and accuracy according to lesion size were also performed.

A total of 59 patients presenting with a suspicious solitary pulmonary nodule < 4 cm in greatest diameter were enrolled at a total of 10 investigative sites. Of these 59 patients, 49 were able for evaluation for efficacy analysis (fulfilled selection criteria, completed technetium (^{99m}Tc) depreotide imaging and histopathological assessment). Of the efficacy population of 49 patients, 46 (94%) presented with an SPN ≤ 3 cm and only 2 of these were < 1 cm. The prevalence of malignancy in the 49 patients was 49% (24 patients). Of the 24 patients positive for malignancy by histopathology, 11 patients (45.8%) had adenocarcinoma, 4 (16.7%) had squamous cell carcinoma, 7 (29.2%) had unspecified NSCLC and 2 (8.3%) had SCLC. Fourteen of the malignancies were graded, mostly Grade 2 (5 of 14) and 3 (7 of 14).

The results of the comparison of the technetium (^{99m}Tc) depreotide scintigraphy read blindly in combination with CT scan and/or chest x-ray relative to the histopathologic diagnosis are summarised in Tables 6, 7,8.

Table 6. Results for All Lesions: Mean (95%CI)

	Diagnostic Performance							
Subgroup	**Accuracy**	**Sensitivity**	**Specificity**	**TP**	**TN**	**FP**	**FN**	**Total**
Majority Blind Read	85.7% (72.1%,93.6 %)	87.5% (66.5%,96.7 %)	84.0% (63.1%,94.7 %)	21	21	4	3	49
Investigator Read	87.8% (74.5%,94.9 %)	91.7% (71.5%,98.5 %)	84.0% (63.1%,94.7 %)	22	21	4	2	49

Table 7. Results for Lesions ≤ 3 cm: Mean (95%CI)

	Diagnostic Performance							
Subgroup	**Accuracy**	**Sensitivity**	**Specificity**	**TP**	**TN**	**FP**	**FN**	**Total**
Majority Blind Read	84.8% (70.5%,93.2 %)	85.7% (62.6%,96.2 %)	84.0% (63.1%,94.7 %)	18	21	4	3	46
Investigator Read	87.0% (73.0%,94.6 %)	90.5% (68.2%,98.3 %)	84.0% (63.1%,94.7 %)	19	21	4	2	46

Table 8. Majority Blind Read, Sample-Based Postitive and Negative Predictive Values: Mean (95%CI)

	Diagnostic Performance	
Subgroup	**PPV**	**NPV**
All Lesions	84.0% (63.1%,94.7%)	87.5% (66.5%,96.7%)
Lesions ≤ 3 cm	81.8% 59.0%,94.0%	87.5% 66.5%,96.7%

The new clinical data from study 829-40 show that there was a total of 31 patients with lesions equal to or less than 2 cm. There were 9 true positives, 18 true negatives, 2 false positives and 2 false negatives. Sensitivity was 81.8%, specificity 90.0% with an agreement rate of 87.1%.

There were 15 patients with nodules greater than 2 cm and equal to or less than 3 cm. In this group, sensitivity was 90.0%, specificity was 60.0% and accuracy 80.0%.

Study 829-40 confirmed that the specificity of technetium (^{99m}Tc) depreotide scintigraphy remains high even in a population with a low prevalence of malignancy. The high negative predictive value demonstrates that technetium (^{99m}Tc) depreotide scintigraphy can, with high confidence, rule out malignancy in this population. The subgroup analysis of the patients with lesions not greater than 3 cm also confirms the value of the test in this population.

A comparison of the results of the 829-40 study with the Phoenix cohort shows that the results of both studies are similar.

In the 829-40 study, the 3 false negatives were an adenocarcinoma (3 cm by CT), an unspecified NSCLC (1.9 cm by CT) and an SCLC (1.0 cm by CT). The 4 false positives were a coccidioidomycosis, 2 cases of pneumonia and an unspecified benign lesion.

In the latter case, although the biopsy result was reported as benign this patient underwent surgical wedge resection of the left lower lobe nodule based on the abnormal technetium (^{99m}Tc) depreotide result The surgical specimen was positive for adenocarcinoma. (Because this result was obtained after the study database was locked, it is not accounted for in the analysis, which assumed this case to be a false positive result).

In the 829-34 trials, the histopathologically positive cases were squamous cell carcinoma (34%), adenocarcinoma (32%), other NSCLC (21%), SCLC (6%) and other malignancies (7%). This is consistent with the common histological types found in patients with primary NSCLC (approximately 30% squamous cell carcinoma, 30% adenocarcinoma and 10% LCLC). Fewer SCLC cases were included than are typically encountered in general practice (about 20%) although clinical and preclinical data suggest that technetium (^{99m}Tc) depreotide scintigraphy can detect SCLC in addition to NSCLC . It is noted that patients were enrolled in the clinical trials evaluating technetium (^{99m}Tc)

depreotide scintigraphy for the detection of lung cancer because there was a suspicion of lung cancer and not because they had known or suspected neuroendocrine cancer.

Efficacy data from the new clinical trial 829-40 of 49 patients has shown that in a population with a prevalence of malignancy of 49%, the accuracy, sensitivity and specificity of technetium (^{99m}Tc) depreotide scintigraphy remains high at 85.7%, 87.5% and 84% respectively with PPV of 84% and NPV of 87.5%. In addition, the negative predictive value of technetium (^{99m}Tc) depreotide scintigraphy remains high in patients with an SPN of size less than 2 to 3cm. The agreement rate between technetium (^{99m}Tc) depreotide scans and histology of 85.7% had a lower 95% confidence limit of 72.1% which exceeded the rate of 70% specified in the protocol at the 0.05 significance level.

Although in study 829-40 it was not possible to indicate the utility of technetium (^{99m}Tc) depreotide in the right lower lobe (the 5 nodules detected in the right lower lobe were all benign), in the pivotal trials (829-34A & 829-34B) the false negative rate for the right lower lobe was the same as for all othe lung regions. (Right lower lobe 13.5%: all other regions 13.2%)

Although the 829-40 protocol did not define an analysis for CT scan alone versus histopathology, some indication of the specificity of the CT (45% spiral CT and 52.7% with contrast) can be obtained from the recent data. Of the 49 patients included in the efficacy patient population, 45 patients had a CT scan performed, and all 45 patient CT scan results were abnormal. Based upon review of the 45 patient CT scan reports, the following data were obtained. These results are relative to the histopathology data:

True positive CT scan reports = 9
True negative CT scan reports = 1
False positive CT scan reports = 8
False negative CT scan reports = 3
Indeterminate CT scan reports = 24

The follow calculations are based on the patients with a determinate CT examination:

CT scan report data alone vs. histopathology:

Sensitivity = TP/TP + FN = 9/9 + 3 = 9/12 =75%

Specificity = TN/TN + FP = 1/1 + 8 = 1/9 = 11%

Accuracy = TP + TN/ TP+TN+FP+FN = 10/ 21 = 48%

PPV = 53%

NPV = 25%

It is noted that all but one SPN were non-calcified lesions.

It appears that the specificity reported for CT is within a wide range, probably depending on several factors but overall the impression is that currently CT scan even with contrast has high sensitivity but low specificity. The ability of low dose spiral CT to achieve high sensitivity and specificity for SPN detection has not yet been demonstrated conclusively. FDG-PET, has a high sensitivity and specificity for characterization of SPNs, with a lower radiation exposure but the technique is expensive and not widely available throughout Europe. The general applicability of MRI for the characterization of SPNs is unknown.

The trials are well designed but with a relatively small number of patients. For evaluating the sensitivity and specificity for the proposed indication of ^{99m}Tc-depreotide scan, histopathological diagnosis of the main presenting lesion has been defined appropriately. In study 829-40 histology was predominantly obtained by fine needle aspiration (FNA) with 5 out of 49 patients having open thoracotomies. Since thoracotomy is considered to be the gold standard (5-8% false negative rate reported for FNA), it is recommended that patients with a negative FNA should be followed up clinically. CT scan and chest X-ray are generally accepted for the primary evaluation and follow-up of lung cancer, and as comparative methods.

Although CT scans are a very sensitive diagnostic tool for patients suspected of lung cancer, the specificity appears to be low, so patients in most cases undergo an invasive procedure to achieve a

final diagnosis. ^{99m}Tc-depreotide scan in conjunction with CT scan or chest x-ray has the same high sensitivity, but the specificity is significantly increased from 11% up to 84% (CI 63.1 - 94.7%) by decreasing the number of false positive and increasing the number of true negatives in patients with SPNs.

In conclusion, these studies provide evidence that ^{99m}Tc-depreotide scan may contribute in the routine clinical setting to differentiate benign from malignant SPNs, when evaluated in conjunction with CT scan or chest X-ray.

Supportive studies

Study 829-30/IIA was a prospective, single centre trial to evaluate ^{99m}Tc-depreotide scan of 12 patients with histological proof of non-small cell lung cancer (NSCLC). The efficacy parameter was the agreement rate between the investigator read ^{99m}Tc-depreotide results and histopathology with respect to presence of disease. ^{99m}Tc-depreotide scans were also compared with FDG-PET results.

One or more lesions were present in ^{99m}Tc-depreotide scan in all the patients. ^{99m}Tc-depreotide results and FDG-PET results agreed in 44 lesions of a total of 54 lesions detected by either modality, while 4 lesions were detected by FDG-PET only, and 6 lesions were detected by ^{99m}Tc-depreotide only.

TNM classification was identical for the two modalities in 10 of 12 cases, but was incomplete in both modalities.

Thus in a small selected group of patients with histological proof of non small cell lung cancer (NSCLC) ^{99m}Tc-depreotide was as efficacious for imaging NSCLC as FDG-PET. However, the number of patients in this study were few and selected, and no firm conclusions can be drawn.

Study 829-22 was a multi-centre, non-randomised, open study conducted in patients having a tumour, known or expected to express SSTR. The aim of the study was to evaluate the ability of ^{99m}Tc-depreotide to detect and localise the tumour sites and to assess its general safety after administration of 10-50 µg peptide labelled with approximately 555 MBq ^{99m}Tc. Early and delayed planar, SPECT and whole body imaging were performed during 3 hours post-injection.

Efficacy was assessed by comparing the scintigraphic results with all other clinically relevant diagnostic and follow-up procedures (CT, biopsy/surgery, MRI, bone scan and other) that contributed to the patient's disease. The investigator at each study centre evaluated the images.

Of a total of 141 patients included, 111 were evaluated for efficacy (only these had all the clinically relevant parameters measured). 28 patients had confirmed or suspected lung cancer and efficacy results were available for 26/28 of these. Scans were correlated to CT in 24 patients, biopsy/surgery in 4 patients and bone scans in 3 patients.

The results of the ^{99m}Tc-depreotide images were in accordance with the confirmatory diagnostic procedures in 20/26 (i.e. 76.9%) patients with proven lung cancer. ^{99m}Tc-depreotide images correlated with CT scans in 18 of 24, and provided additional information in all 6 cases were the two modalities did not correlate.

For early and delayed imaging performed within the 3 hours post-injection period, no appreciable difference was reported. In general, the image quality was better for the delayed images (i.e. 90 minutes post-injection).

Two additional studies (P829-30A & P820-30B) were identical with respect to aims, study design and procedures, patient population, efficacy and safety assessments, and analyses. The aims of these studies (uncontrolled and not randomised) were to evaluate the ability of ^{99m}Tc-depreotide to detect and localise somatostatin receptor expressing neuroendocrine tumours as compared to ^{111}In pentreotide (OctreoScan) and to evaluate the general safety of ^{99m}Tc-depreotide scintigraphy. Blinded reads were performed by three independent nuclear medicine physicians, and were compared to the final institutional diagnosis (FIC), which was considered definitive.

Of the 252 patients included in the studies, 34 patients had confirmed or suspected primary neuroendocrine lung tumours, and these patients were evaluated for efficacy. Efficacy was evaluated by comparing the agreement rate, sensitivity and specificity of ^{99m}Tc-depreotide and ^{111}In pentreotide using FIC diagnosis as reference method.

For the 34 patients the agreement rate for both blind read and (investigator read) technetium (^{99m} Tc) depreotide was 79% (27/34), the sensitivity 82% (23/28) and the specificity 66% (4/6). Agreement rate, sensitivity and specificity were all higher (not statistically different) for indium (^{111} In) pentetreotide.

Discussion on clinical efficacy

The number of subjects in the pivotal studies is relatively small however these were well conducted. The statistical methodology was changed after study commencement because a cut-off point was chosen based upon an observed agreement rate between CT scan results and histopathology results to be no higher than 65% as documented in the literature.

Technetium (^{99m} Tc) depreotide images for the combined pivotal studies in patients with suspected lung tumours resulted in an agreement rate that was significantly higher than 70%, a figure chosen because of the literature agreement rates between CT and histopathology. Sensitivity at 83.7% was less than CT and specificity at 59.5% greater than CT. While the specificity is far from ideal even high values for both sensitivity and specificity are of limited value if the predictive value is not appropriate. The calculated PV and PPV indicate that at a disease prevalence of 30-50% a negative ^{99m}Tc-depreotide scan has a high probability of correctly diagnosing non-malignancy while a positive scan has a lower probability of correctly predicting a malignancy. However the overall population for which technetium (^{99m}Tc) depreotide is proposed will have a higher than 50% prevalence of malignancy. In a large proportion of these a negative technetium (^{99m}Tc) depreotide scan would still have to be followed by biopsy.

Patients with SPN are often a diagnostic challenge. While the prevalence of malignancy was high (79%) in the SPN population, in study 829-P in many reported studies only 28-39% of resected SPNs are malignant. In all subgroups of SPN, technetium (^{99m}Tc) depreotide used in conjunction with chest X-ray or CT provided improved agreement rate and specificity while retaining sensitivity > 88.9%, compared to technetium (^{99m}Tc) depreotide or CT alone.

Results appear to be similar to the reported results of FDG-PET imaging of SPN patients with a sensitivity of 96% and a specificity of 69%. FDG-PET has a high sensitivity and specificity with a lower radiation dose than technetium (^{99m}Tc) depreotide but is not widely available throughout Europe.

The results suggest that including technetium (^{99m}Tc) depreotide in the evaluation of SPNs (in addition to chest X-ray or CT scan) could reduce the number of false positives and the number of invasive procedures necessary to rule out malignancy. There are fewer false positive results when including technetium (^{99m}Tc) depreotide in the diagnostic work-up. A negative diagnosis would suggest that biopsy could be withheld in those patients with low likelihood of malignancy and a repeat chest X-ray or CT scan performed to assess change at an appropriate time point. It appears therefore that in the subgroup of patients with SPNs technetium (^{99m}Tc) depreotide may be a useful imaging agent.

In a recent clinical study with a prevalence of malignancy of 49%, the PPV for NeoSpect in association with CT/chest X-ray was 84% (CI 63.1-94.7%) for all SPNs and 81.8% for lesions equal to or less than 3cm. The negative predictive value was 87.5% (CI 66.5-96.7%) for all lesions and 87.5% for lesions equal to or less than 3cm. However, histology was predominantly obtained by fine needle aspiration (FNA) with 5 out of 49 patients having open thoracotomies. In view of the false negative rate of FNA (5-8% false negative rate reported), thoracotomy is considered to be the gold standard. Patients with a negative FNA should be followed up clinically as some FNA biopsies may give false negative results

Clinical safety

Patient exposure

A total of 970 individuals were included in 14 clinical studies. A total of 968 of these patients were exposed to the study agent (58 volunteers and 910 patients); two patients withdrew consent prior to injection of ^{99m}Tc-depreotide. Most subjects received a single injection but 12 patients received 2 and one patient 3 injections. 825 (90.8%) of subjects received the formulation intended for commercial use.

151 patients were evaluated for safety only. (These included normal volunteers, patients with neuroendocrine tumours, lymphoma and melanoma).

Of the 968 subjects (514 males and 454 females) exposed to the study agent 835 completed all assessments.

The dose of the peptide was approximately 50 µg in 11 of the 14 clinical studies, whereas in 3 studies the dose ranged from 5 to 50 µg (829-20, 829-22, 829-23). The activity of ^{99m}Tc ranged from 296-740 MBq, but was 555-740 MBq in most of the studies.

Safety parameters assessed included exposure to the study agent, prior and concomitant medications, adverse events, clinical laboratory tests, vital signs, immunogenicity of the study agent, and post-injection observation time of 24 hours. All adverse events were coded and classified using the WHO-ART dictionary or COSTART.

Adverse events

There were no deaths or other serious events reported in any of the 14 clinical studies, and no patients or healthy volunteers discontinued the study due to adverse events.

A total of 52 subjects reported 79 adverse events. Of these 17 events were considered not related to ^{99m}Tc-depreotide. The most common event was headache (8 events), nausea and diarrhoea (6 events each). The majority of all events (Table 9) were judged to be mild (57 events) or moderate by the investigators.

Eight events in 4 patients (< 1%) were judged to be severe (headache, backache (twice), lower extremity pain/severe diarrhoea and abdominal pain/severe back pain/toothache). However, all events were judged as probably not related to administration of ^{99m}Tc-depreotide.

Table 9. Incidence of Adverse Events

Body System / Symptom	Injections total n=923 No of events:
Body as a Whole	20 (2%)
Gastrointestinal	19 (2%)
Central and Peripheral Nervous System	15 (< 2%)
Cardiovascular	5 (< 1%)
Respiratory	3 (< 1%)
Special Senses	3 (< 1%)
Psychiatric	2 (< 1%)
Endocrine	1 (< 1%)
Metabolic and Nutritional	1 (< 1%)
Musculo-skeletal	1 (< 1%)
Platelet, Bleeding and Clotting	1 (< 1%)
Resistance Mechanism	1 (< 1%)
Skin and Appendages	1 (< 1%)
Vision	1 (< 1%)
White Cell and RES	1 (< 1%)
Headache	11 (<1%)
Nausea	6 (< 1%)
Diarrhoea	6 (< 1%)
Dizziness	4 (< 1%)
Back Pain	5 (< 1%)
Fatigue	3 (< 1%)
Vasodilatation	3 (< 1%)
Vomiting	3 (< 1%)

Flushing	3 (< 1%)
Taste Perversion	3 (< 1%)
Abdominal Pain	2 (< 1%)
Chest Pain	2 (< 1%)
Somnolence	2 (< 1%)
Hypertension	2 (< 1%)
Pain/discomfort	2 (< 1%)
Pain/toothache	2 (< 1%)

In study 829-34A and 829-34B, 237 of 271 patients (87%) received concomitant medications, mainly calcium channel blockers, diuretics and corticosteroids. As so few adverse events were reported during the studies (5% or fewer of patients in any study grouping) adverse events were not subgrouped by specific medications.

A slightly higher incidence of adverse events was reported in subgroup patients with abnormal renal and hepatic function (n = 10) compared to patients with normal function. A comparison of specific adverse events was not possible because of the small number of patients and events in this subgroup.

No adverse events were reported in the 13 patients (12x2 and 1x3) who received multiple injections.

Clinical laboratory values

Clinical laboratory values were assessed in 11 of the 14 clinical studies with 99mTc-depreotide.

The mean changes from pre-injection (n=737 patients) to each post-injection evaluation (at 1 hour n=224 patients; at 2-6 hours n=552 patients and at 18-30 hours n=674 patients) in clinical chemistry for all parameters (i.e. alkaline phosphatase, AST, ALT, LDH, total protein, total bilirubin, BUN and creatinine) were small. The same were reported in mean changes for all hematology parameters (i.e. haematocrit, haemoglobin, RBW, WBC, platelets). No clinically significant changes were observed in any urinary parameters as well.

Clinically significant changes defined as any 25% change from the pre-injection value (in both directions) were found in a few patients only and these are summarised in Table 10.

Table 10. Significant changes from preinjection laboratory values (i.e., any 25% change from the pre-injection value in both directions)

Parameter	Patients
Alkaline phosphatase	1/661 (<1%)
AST	3/670 (<1%)
ALT	5/670 (<1%)
LDH	2/652 (<1%)
Total protein	1/630 (<1%)
Total bilirubin	1/667 (<1%)
BUN	4/674 (<1%)
Creatinine	0/673 (0%)
Hematocrit	2/636 (<1%)
Haemoglobin	2/635 (<1%)
RBC	2/635 (<1%)
WBC	8/607 (1%)
Neutrophils	2/611 (<1%)
Basophils	3/617 (<1%)
Eosinophils	8/618 (1%)
Monocytes	1/619 (<1%)
Platelets	1/589 (<1%)

In study 829-22 it is noted that some patients with normal plasma glucose pre-injection had high values post-injection. Patients (n = 12) with high values pre-injection rose substantially post-injection. Some low values of glucose were recorded post-injection.

One slightly high calcium result was recorded post-injection.

Immunogenicity

Analysis of IgG and IgM demonstrated no antibodies against depreotide (study 829-12 volume reference, study 829-13 volume reference) in 18/32 patients in whom pre- and post-injection (3 weeks) serum was available.

Vital signs

Systolic and diastolic blood pressures, pulse and respiration rates were assessed in 12 of 14 clinical studies. However, none of 6 cases deemed noteworthy were considered to be associated with the administration of ^{99m}Tc-depreotide.

Discussion on clinical safety

Assessment of safety parameters has been performed according to standard criteria for radiopharmaceuticals, and the results indicate that no serious reactions are to be expected after intravenous bolus injection of ^{99m}Tc-depreotide. The dose of technetium (^{99m}Tc) 15-20 mCi advocated is not inconsiderable but compares with other, approved radiopharmaceuticals. It is thought to be safe with no abnormally high absorption by any organ. The probability of radiation-induced toxicity from ^{99m}Tc-depreotide is therefore not considered to be high.

5. Overall conclusions, benefit/risk assessment and recommendation

Quality

The quality of this product is considered to be acceptable when used in accordance with the conditions defined in the SPC. Physicochemical and biological aspects relevant to the uniform clinical performance of the product have been investigated and are controlled in a satisfactory way. Some CPMP Members noted the rather high radiochemical impurity levels in the product. However, this concern was not shared by the majority of CPMP Members and the applicant has initiated further steps to address these concerns and provided a written undertaking that these concerns will be fully addressed (as a follow-up measure) and the data supplied within a specified timeframe.

Although a few minor quality points remain to be fully clarified, the remaining data (for example, further batch analyses data) do not raise any major concerns and would not prevent authorisation of the product. These outstanding data will be provided post-authorisation (as follow-up measures) within a specified timeframe.

Preclinical pharmacology and toxicology

Overall the primary pharmacodynamic and toxicology studies provided adequate evidence that ^{99m}TC-depreotide has the appropriate properties for the intended application. Although ^{99m}Tc-depreotide is distributed to various organs and tissues in the human, it is labelled with ^{99m}Tc which has a short physical half-life (6.0 hours) and emits only low/medium energy gamma photons and low-energy Auger and conversion electrons. The absorbed radiation doses to the various organs and tissues in the human from a 740 MBq dose of ^{99m}Tc-depreotide are rather high in comparison with other, approved radio-pharmaceuticals, but the probability of radiation induced toxicity from ^{99m}Tc-depreotide is not considered to be high.

The compound accumulated in renally impaired animals. Although the human data indicate that there is no need to reduce the dose in renally impaired patients, this has been reflected in the SPC.

Efficacy

In all subgroups of SPN, technetium (^{99m}Tc) depreotide used in conjunction with a chest X-ray or CT scan showed an agreement rate with histology significantly higher than 70% ($P<0.05$).

The prevalence of malignancy was high (79%) in the SPN population in the pivotal studies 829-34A & B. In most reported studies 28-39% of resected SPNs are malignant and the SPN is much less likely to be malignant in people younger than 35 years.

An additional clinical study 829-40 has shown that, using ^{99m}Tc-depreotide in combination with an X-ray or CT scan in a 49 patient population with a prevalence of malignancy of 49%, the accuracy, sensitivity and specificity of technetium (^{99m}Tc) depreotide scintigraphy remains relatively high. The negative predictive value of technetium (^{99m}Tc) depreotide scintigraphy was 87.5% in patients with an SPN of size less than 3cm. For lesions less than 3cm there was a total of 31 patients with lesions equal to or less than 2 cm. Of these there were 9 true positives, 18 true negatives, 2 false positives and 2 false negatives. Sensitivity was 81.8%, specificity 90.0% with an agreement rate of 87.1%. There were 15 patients with nodules greater than 2 cm and equal to or less than 3 cm. In this group, sensitivity was 90.0%, specificity was 60.0% and accuracy 80.0%. In this study histology was predominantly obtained by fine needle aspiration (FNA) with 5 out of 49 patients having open thoracotomies. Since thoracotomy is considered to be the gold standard (5-8% false negative rate reported for FNA), it is recommended that patients with a negative FNA should be followed up clinically.

CT scan and chest X-ray are generally accepted for the primary evaluation and follow-up of lung cancer, and as comparative methods.

The results suggest that including technetium (^{99m}Tc) depreotide in the evaluation of SPNs (in addition to chest X-ray or CT scan) could reduce the number of false positives and the number of invasive procedures necessary to rule out malignancy.

The results presented in the initial NeoSpect application were obtained in prospective trials in which the CT scans were performed with contrast enhancement in 71 % of the SPN patients. Whether contrast was used or not was at the discretion of the radiologist. The CT scans in the 829-34 trials were thus largely results obtained with contrast enhancement and showed a specificity of 7%. In study 829-40 spiral CT was used in 45% of patients and contrast was used in 52.7% of patients. The specificity reported for CT is within a wide range, probably depending on several factors but the currently widely accepted position of CT scan is that it has high sensitivity but low specificity. In addition there is insufficient evidence to conclude, and in particular, no controlled study showing definitively, that spiral CT scan has adequate specificity and sensitivity to predict with conviction benign vs. malignant SPNs.

FDG-PET has a sensitivity and specificity for characterization of SPNs comparable to those of NeoSpect but with lower radiation exposure.

The general applicability of using MRI for the characterisation of SPNs is unknown.

It appears therefore that in the subgroup of patients with SPNs technetium (^{99m}Tc) depreotide may be a useful imaging agent. While the dose of technetium (^{99m}Tc) advocated (740 MBq dose) is not inconsiderable, it is thought to be safe with no abnormally high absorption by any organ. The proposed indication for NeoSpect is scintigraphic imaging of suspected malignant SPNs in combination with CT scan or chest X-ray. The radiation dose from NeoSpect alone does not appear to pose a safety risk for radiation-induced injury to tissues. Following the addition of the radiation dose from a CT scan, the risk still remains within the accepted limits. Using spiral CT instead of conventional CT however the radiation dose may be kept to a minimum.

Overall the benefit/risk ratio is positive for the use of NeoSpect for scintigraphy of solitary pulmonary nodules (SPNs) in association with chest X-ray or CT scan.

Safety

Toxicity studies indicate a good safety profile for ^{99m}Tc-depreotide. No organ specific toxicity was identified, the product is non-genotoxic, non-antigenic and non-irritant.

Secondary pharmacology, drug interaction, reproductive toxicity and carcinogenicity testing were not included in the dossier, considering the proposed indication, these omissions are justified. A detailed environmental risk assessment (ERA) was not submitted but the annual use in the EU is predicted to be <1g ^{99m}Tc-depreotide. Predicted environmental concentrations (PECs) would therefore be well below the action limit of 0.01 µg/L.

Benefit/risk assessment

The chemistry and pharmacy data supporting the application for NeoSpect required supplementation in several areas, in particular tightening of the limits in the specifications for both the active ingredient and the finished product (both before and after reconstitution). Although the specifications have now been amended and are satisfactory to ensure the quality of the product, some validation data remains to be provided, but the applicant has confirmed that these data will be provided as follow-up measures. Some Members held a divergent position on the pharmaceutical quality, in particular the high level of radiochemical impurities. However, this concern was not shared by the majority of CPMP Members who were of the opinion that the toxicology of these impurities was not in question bearing in mind the low dose, and that there was no evidence of a negative effect on the clinical performance of the product, as justified in Part IV.

The safety profile ^{99m}Tc-depreotide is in accordance with that expected from pre-clinical studies. Adverse effects were uncommon and of mild intensity. Exposure to ionising radiation was adequately addressed. No major safety concerns have emerged from the clinical studies.

During the evaluation of this product there were concerns over the clinical usefulness. Additional data were provided which show that in the subgroup of patients with SPNs that ^{99m}Tc-depreotide may be a useful imaging agent. In all subgroups of SPN, ^{99m}Tc-depreotide used in conjunction with a chest X-ray or CT scan showed an agreement rate with histology significantly higher than 70% ($P<0.05$).

^{18} FDG-PET has high sensitivity and specificity and uses a lower radiation dose than ^{99m}Tc-depreotide, however it is not widely available throughout Europe.

Thus, the CPMP considered that the efficacy of NeoSpect in the diagnosis (by scintigraphic imaging) of suspected malignant tumours in the lung, in combination with a CT scan or chest X-ray, in patients with solitary pulmonary nodules was acceptable.

Recommendation

Based on the CPMP review of data on quality, safety and efficacy, the CPMP considered by majority decision that the benefit/risk profile of NeoSpect in the diagnosis by scintigraphic imaging of suspected malignant tumours in the lung after initial detection, in combination with CT scan or chest X-ray, in patients with solitary pulmonary nodules, was favourable and therefore recommended the granting of the marketing authorisation.