2.4 Nonclinical Overview

Product name:
TERIPARATIDE
20 micrograms/80 microliters
solution for injection in pre-filled pen

Drug Substance:
Teriparatide

Dosage Form, Strength:
Solution for injection, 20 µg/80 µL

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List of abbreviations

^{125}I	Radioisotope of Iodine
AP	Alkaline Phosphatase
ATC	Anatomical Therapeutic Chemical Classification
AUC	Area Under the Concentration-Time Curve
BfArM	Federal Institute for Drugs and Medical Devices (Bundesinstitut für Arzneimittel und Medizinprodukte)
BMC	Body Mineral Content
BMD	Bone Mineral Density
BMI	Body Mass Index
CASRN	Chemical Abstracts Service Registration Number
C_{max}	Maximum concentration
DNA	Deoxyribonucleic Acid
E. coli	Escherichia coli
e.g.	*exempli gratia* (for example)
EPAR	European Public Assessment Report
FDA	Food and Drug Administration (US)
GIO	Glucocorticoid-Induced Osteoporosis
GLP	Good Laboratory Practice
Gla-OC	Carboxylated-Type of Osteocalcin
GRAS	Generally Recognised As Safe
HSDB	Hazardous Substances Data Bank
i.e.	*id est* (that means)
IGF	Insulin-Like Growth Factor
INN	International Nonproprietary Name
i.v.	Intravenous(ly)
LPS	Lipopolysaccharide
mRNA	Messenger Ribonucleic Acid
MEDLINE	Medical Literature Analysis and Retrieval System Online
MPS	Methylprednisolone
NOEL	No-Observed-Effect Level
NS	Normal Saline
NTX	Amino-Terminal Cross-Linked Telopeptide Type 1 Collagen
OC	Osteocalcin
OPG	Osteoprotegerin

OVX	Ovariectomised
PTH	Parathyroid Hormone
Ph. Eur.	Pharmacopoeia Europaea
PubMED	Public MEDLINE
QCT	Quantitative Computed Tomography
r	Correlation Coefficient
RANK	Nuclear Factor Kappa-B
RANKL	Nuclear Factor Kappa-B Ligand
s.c.	Subcutaneous(ly)
SCOGS	Select Committee on GRAS Substances
SERM	Selective Oestrogen Receptor Modulator
SmPC	Summary of Product Characteristics
TCP	Tricalcium Phosphate
TGF	Transforming Growth Factor
TOXNET	Toxicology Data Network
vs.	Versus

2.4.1. Overview of the nonclinical testing strategy

2.4.1.1. Introduction

This Nonclinical Overview provides a comprehensive bibliographic review of relevant published references of teriparatide in order to establish the acceptable level of safety and efficacy of this active agent from a nonclinical point of view. With this application, the applicant is seeking approval of the product TERIPARATIDE 20 micrograms/80 microliters solution for injection in pre-filled pen, which contains teriparatide (20 µg per 80 µL or 250 µg per mL). The legal basis for this application refers to Article 10(1) of Directive 2001/83/EC - relating to applications for generic medicinal products.

The document aims to provide concise and up-to-date information on teriparatide as well as on the excipients used for the solution and of the impurities which may be present in the product, if applicable. The overview addresses the recently published literature (MEDLINE via PubMED and TOXNET; up to July 2018) allowing for any new information on the pharmacology and toxicology of the active agent to be taken into account. Thereby, the overview demonstrates that the proposed summary of product characteristics (SmPC, G1) adequately reflects the prevailing scientific knowledge on teriparatide in the indications claimed by the applicant.

Teriparatide is the 1-34 N-terminal fragment of endogenous human parathyroid hormone (Figure 1); the peptide can be either produced in *Escherichia coli (E. coli)* using recombinant DNA technology or chemically synthesised by solid phase peptide synthesis. The mode of synthesis does affect neither the preclinical properties nor the clinical performance of teriparatide. The theoretical monoisotopic mass of teriparatide is 4115.1309 Dalton ( ). The amino acid sequence is: [1].

TERIPARATIDE contains teriparatide manufactured by chemical solid phase peptide synthesis whereas teriparatide in the reference medicinal product Forsteo® is of recombinant origin and produced in *E. coli*. The preclinical and clinical characteristics of teriparatide are irrespective of the mode of synthesis, and products containing recombinant as well as chemically synthesised teriparatide have been approved in the European Union (G2, G3, G4).

[1]

Figure 1: 2D structure of teriparatide (G6)

The product under discussion contains teriparatide as active substance. The product is presented as a colourless, clear solution, containing teriparatide (250 µg per mL) in a prefilled pen. Each dose of 80 µL contains 20 µg teriparatide. Besides teriparatide, the ingredients

All excipients are commonly used in the preparation of injectables and comply with European Pharmacopoeia requirements.

Table 1: Composition of the TERIPARATIDE 20 micrograms/80 microliters solution for injection in pre-filled pen

	Name of the ingredient	Unit/mL	Function	Reference to Standards
1	Teriparatide			In-house Monograph
2	Glacial acetic acid			Ph. Eur.[2]
3	Anhydrous sodium acetate			Ph. Eur.[2]
4	D-Mannitol			Ph. Eur.[2]
5	Metacresol			Ph. Eur.[2]
6	Hydrochloric acid	.		Ph. Eur.[2]
7	Sodium hydroxide			Ph. Eur.[2]
8	Water for injections			Ph. Eur.[2]

q.s. - *quantum satis*

[2]Current edition

2.4.1.2. Targeted indication

The product TERIPARATIDE is indicated for treatment of osteoporosis in postmenopausal women and in adult men at increased risk of fracture as well as for treatment of osteoporosis associated with sustained systemic glucocorticoid therapy in adult women and men at increased risk for fracture (for details see G1).

As a truncated form of the human parathyroid hormone, teriparatide belongs to the pharmaco-therapeutic group "Calcium homeostasis, parathyroid hormones and analogues (Anatomical Therapeutic Chemical Classification System (ATC) Code H05AA02)".

2.4.1.3. GLP Status of the nonclinical studies

The development of teriparatide is closely related to the understanding of bone biology and the pathogenesis of osteoporosis. The important role of oestrogen in regulating bone metabolism in women (and in men) was initially described in the 1940s. In the 1980s, it was demonstrated that oestrogen replacement and possibly the administration of calcitonin along with calcium and vitamin D supplementation benefited postmenopausal woman suffering from osteoporosis. Findings from the placebo-controlled Women's Health Initiative published in the early 2000s demonstrated that, while hormone replacement therapy can delay or improve osteoporosis and decrease the risk for fragility fractures, it increases the risk of cardiovascular diseases and breast cancer. Accordingly, alternative treatment approaches mimicking the antiresorptive activity of oestrogen but with better tolerability were developed over the last decades. Currently, bisphosphonates are the most widely used drugs for the prevention and treatment of osteoporosis (13, 24).

The development of teriparatide can be ascribed to early observations indicating that in mammals, parathyroid hormone (PTH), a peptide of 84 amino acids, controls bone metabolism and that hyperparathyroidism leads to marked bone loss due to increased bone resorption. In patients with hypoparathyroidism, in contrast to continuous exposure, intermittent therapy with PTH, increased bone formation with smaller increases in bone resorption, resulting in a net anabolic effect. A similar response was observed with the subcutaneous application of a PTH analogue comprising the first 34 amino acids of the endogenous hormone (13, 15). Teriparatide was developed in the 1990s by Ely Lilly and Company and introduced in the pharmaceutical market in the early 2000s as the first anti-osteoporotic agent with anabolic activity.

It may be assumed that the preclinical development of teriparatide was predominantly done in a systematic way and were thus performed in compliance with good laboratory practice (GLP). However, even if not all studies are consistent with the GLP requirements under all issues demanded, this is not expected to affect the overall conclusions of this Nonclinical Overview.

2.4.1.4 Impurities and degradants

The product TERIPARATIDE contains teriparatide (20 µg per 80-µl dose or 250 µg per mL) as the active pharmaceutical ingredient. The product also contains a number of excipients (see also sections 2.4.1.1 and 2.4.5), which all are of the appropriate pharmaceutical grade and conform to

the respective monographs of the European Pharmacopoeia (Ph. Eur., for details see Module 3.2.P.4.1).
Based on the available chemical and pharmaceutical data, the excipients used in the present formulation are unlikely to pose any significant toxicity hazard to humans in the amounts present in the formulation, when administered parenterally at recommended doses.

Although some differences were found in the impurity profile of Forsteo and teriparatide 20 µg/80 µl solution for injection in pre-filled pen, global purity profile can be considered similar for both formulations. Three individual impurities are found in teriparatide 20 µg/80 µl solution for injection in pre-filled pen batches that are not detected in Forsteo.
These impurities not being present in the formulation of reference product are in all cases below the identification threshold (>0.5%) defined in the Ph Eur current edition monograph 2034 for peptides obtained by chemical synthesis, and well below the qualification threshold (>1.0%) stated in the mentioned monograph (for details see Module 3.2.P.2.2.2.2).

The comparison of degradation profile of samples of Forsteo and teriparatide 20 µg/80 µl solution for injection in pre-filled pen stored for 30 days at 40±2ºC // 75±5% RH showed that both formulations show the same degradation profile.
This confirms that the origin of the drug substance (either recombinant or chemical synthesis) has no impact at all on the degradation profile of the product.

2.4.2 Pharmacology

2.4.2.1 Introduction

In mammals, parathyroid hormone (PTH) is synthesised and stored in the chief cells of the parathyroid glands. Synthesis is regulated by a feedback mechanism involving the level of blood calcium (and, to a lesser degree, that of magnesium). The primary function of PTH is to control calcium concentration in the extracellular fluid, which it does by affecting the rate of transfer of calcium into and out of bone, resorption in the kidneys, and absorption from the gastrointestinal tract. The major initial effect on bone is to mobilise calcium from the bone to the extracellular fluid; later, bone formation may be enhanced. Besides PTH, vitamin D and calcitonin are major hormones in the maintenance of calcium homeostasis.

Human PTH is an 84-amino acid peptide; teriparatide is the 1-34 N-terminal fragment of PTH that appears to contain all the anabolic properties of the full-length peptide. Both teriparatide and endogenous PTH mediate their biological effects via specific, high-affinity membrane cell-surface receptors expressed on osteoblasts and renal tubular cells. Both molecules bind to the receptors with the same affinity and exert the same physiological effects on bone and kidney (3).

Osteoporosis is a largely asymptomatic, widespread disorder that affects more than 75 million people throughout Europe, the US and Japan, and its incidence is likely to increase as life expectancy rises. Older age, high body mass index (BMI), smoking and a sedentary lifestyle are recognised risk factors for osteoporosis; the same applies to long-term glucocorticoid therapy. Osteoporosis is characterised by low bone mineral density (BMD), which leads to decreased bone strength and consequently an increased risk of fragility fractures (3).

2.4.2.2 Pharmacodynamic effects and mode of action

Of note, teriparatide can be produced using DNA technology and extraction of the fragment from a transformed non-pathogenic *Escherichia coli* (*E. coli*) K12 strain (7). An alternative approach is chemical manufacturing using solid phase peptide synthesis resulting in a molecule consisting of 34 amino acids. With both, irrespective of the mode of production, there are no posttranslational modifications. Both peptides do, without any chemical modification. The latter method has been chosen for teriparatide contained in the product under discussion.

Mode of action
Since teriparatide and the 34 N-terminal amino acids of PTH bind to the same receptors with the same affinity, it can be assumed that they produce the same physiological effects on bones and kidneys (5, 7, 10).

Parathyroid hormone is a major regulator of calcium homeostasis that developed millions of years ago when evolution moved organisms away from the calcium-rich environment of the sea onto the continents that offered only little calcium supply. A decrease in serum calcium level stimulates PTH release from the parathyroid glands and triggers a variety of biological responses that help to maintain calcium homeostasis under calcium-depleted conditions. One such important response is calcium mobilisation from the skeleton, but there is also the stimulation of 1-hydroxylase activity in the kidneys, leading to increases in serum 1,25-dihydroxyvitamin D levels. This step promotes calcium absorption in the gastrointestinal tract. In the distal tubule in the kidneys, PTH enhances the reabsorption of filtered calcium (7, 10).

The primary target cell for PTH, and accordingly for teriparatide, in bone is the osteoblast. Activation of specific receptors stimulates several cascades including the adenylyl cyclase activating G-protein-coupled protein. The anabolic response is elicited by the activation of protein kinases A and C. The final response to intermittent PTH stimulation involves the induction of several growth factor genes, such as insulin-like growth factors (IGFs) and transforming growth factor (TGF). Besides the IGF system, the receptor activator of nuclear factor kappa-B / osteoprotegerin (RANKL / OPG) system convey the differential responses of bone cells towards PTH (7).

Both under *in vitro* as well as in *vivo* conditions, it has been shown that stimulation of RANKL decreases OPG mRNA and protein expression. RANKL is a potent stimulator of osteoclast proliferation and activity (7, 9). In cultured marrow cells from 6-week old C57BL/6 mice, Locklin et al. (2003) demonstrated that 4 days of intermittent PTH treatment increased mRNA for osteoblast differentiation markers, alkaline phosphatase (AP) and type I procollagen whereas continuous treatment resulted in production of large numbers of multinucleated osteoclasts. Although IGF-I mRNA did not increase after intermittent treatment, it was consistently higher than after continuous treatment, and the addition of anti-IGF-I neutralising antibodies prevented the increase in bone formation indices observed with intermittent treatment. By contrast, after continuous treatment, gene expression of RANKL was increased and that of OPG was decreased, resulting in a 25-fold increase in the RANKL / OPG ratio. According to the authors, in this model system, intermittent PTH treatment enhances osteoblast differentiation through an IGF-I dependent mechanism and continuous PTH treatment enhances osteoclastogenesis through reciprocal increases in RANKL and decreases in OPG (16).

In summary, the pattern of administration defines the action on the skeleton: Whereas continuous teriparatide exposure accelerates bone resorption via enhanced osteoclast activity, intermittent exposure stimulates bone formation. One of the first cellular responses that can be seen within days with intermittent teriparatide treatment in rats is a transformation of resting bone lining cells to cells that resemble mature osteoblasts and carry the typical endoplasmatic reticulum seen in light and electron microscopic studies (7).

Pharmacodynamics

When teriparatide is administered by once-daily subcutaneous (s.c.) injection to rodents, monkeys or humans, bone mass is increased due to an increased number of osteoblasts and bone formation rate. The cause of the increased number of osteoblasts after teriparatide administration has been studied extensively. Teriparatide suppresses osteoblast apoptosis and activates dormant bone-lining cells so that they become active osteoblasts. Both of these actions of teriparatide involve postmitotic, mature cells of the osteoblast lineage. It is less clear whether teriparatide also acts on early osteoprogenitors to increase the number of new osteoblasts (2, 10).

Balani et al. (2017) investigated the role of osteoblast precursors participating in the increase in osteoblast numbers after administration of teriparatide. In addition, the fate of such cells after cessation of teriparatide administration was assessed. The authors employed a lineage-tracing strategy that uses transgenic mice expressing Sox9-creERT2, R26RTomato reporter to study the role of osteoblast precursors. In this model, teriparatide increased the numbers of osteoblast precursors and drove their differentiation into mature osteoblasts. Unexpectedly, following withdrawal of teriparatide therapy, bone marrow adipocytes increased significantly in number. Some of these adipocytes derived from cells marked by Sox9-cre expression weeks earlier. Continued therapy with teriparatide prevented the appearance of adipocytes. Selective, inducible deletion of the PTH receptor in Sox9-cre cells demonstrated that PTH receptor expression is required for teriparatide-mediated increases in early osteoblast precursors. The increase in early precursors after teriparatide administration was associated with robust suppression of precursor apoptosis without affecting their rate of proliferation. Thus, teriparatide increases the numbers of early cells of the osteoblast lineage, hastens their differentiation into osteoblasts, and suppresses their differentiation into adipocytes *in vivo* (2).

In the 1990s, a number of preclinical studies demonstrated that intermittent treatment with PTH increases osteoblast number and bone formation in growing and adult rats. Animal models for postmenopausal osteoporosis are generated by ovariectomy. Bone loss occurs in states of oestrogen deficiency due to enhanced bone resorption and impaired osteoblast function. However, there are important differences in skeletal physiology between rodents and humans. While the human skeleton is dominated by osteonal remodelling, the rat skeleton is dominated by skeletal growth modelling, resulting in dissimilar effects when exposed to teriparatide (19). Although species such as rabbits and monkeys are remodelling species, their bone physiology differs in details from that seen in humans. Therefore, these animal models do not correspond to the characteristics of postmenopausal osteoporosis in women and findings on the pharmacology of teriparatide gained in these models only partially translate into humans.

In animal models, it was demonstrated that teriparatide increases the structural integrity of trabecular bone and it increases bone strength. The anabolic effects are pronounced in the trabecular bone and on the endosteal surface of cortical bone. In rats, cortical bone mass and strength were increased. In remodelling species such as monkeys and rabbits, PTH and teriparatide increase cortical bone porosity; however, due to the concomitant increase in bone diameter (i.e. an

anabolic effect on the periosteal surface), bone strength seems to be unaffected by the increased porosity (10).

Animal studies performed before the authorisation of the originator product Forsteo demonstrated that intermittent subcutaneous (s.c.) administration of teriparatide stimulated bone apposition and new bone formation in rats, mice, rabbits and monkeys. In ovariectomised (OVX) monkeys, teriparatide administered at doses of 1 and 5 µg/kg per day for 18 months improved bone mass, architecture and mechanical strength of trabecular and cortical bone without producing any adverse effects. In osteopenic OVX rats, the minimally efficacious dose was between 0.3 and 1.0 µg/kg per day. After treatment withdrawal in monkeys, beneficial changes in bone mass and biomechanical properties were sustained for about 6 months. Overall, teriparatide stimulated both modelling and remodelling of bone leading to improved bone quality (G2).
In OVX cynomolgus monkeys, intermittently administered teriparatide increased intracortical bone turnover and porosity without reducing bone strength in the humerus (4). Chen et al. (2007) aimed to analyse relationships between iliac crest bone microarchitecture and bone microarchitecture and strength in the vertebrae and femoral neck after 18 months of teriparatide treatment in OVX cynomolgus monkeys. The authors studied the relationships between trabecular and cortical bone spatial architecture of the lumbar vertebra and femoral neck and bone strength. They aimed to ascertain how static histomorphometry of the iliac crest correlates with vertebral and femoral neck strength and to assess how 6-month iliac crest biopsy parameters compared with 15-month biopsy parameters as predictors of vertebral and femoral neck strength and architecture after 18 months of teriparatide treatment. OVX monkeys were administered vehicle (n = 20), teriparatide 1.0 µg/kg/day (n = 19) or teriparatide 5.0 µg/kg/day (n = 21) for 18 months. Iliac crest biopsies were obtained at 6 and 15 months after initiation of treatment. Animals were killed after 18 months of treatment, and adjacent vertebrae or contralateral proximal femora were processed for biomechanical or histomorphometric analyses. Static histomorphometric parameters of the 6- and 15-month biopsies were significantly correlated with the vertebral and femoral neck parameters obtained at 18 months of teriparatide treatment (Table 2). Iliac crest biopsy parameters at 6 and 15 months also correlated with vertebral and femoral neck strength at 18 months. Static histomorphometry of the lumbar vertebra and femoral neck at 18 months also significantly correlated with strength at these sites. However, cortical bone volume of the lumbar vertebrae had the strongest correlation with vertebral and femoral neck strength (r = 0.74 and 0.71, respectively). The authors concluded that teriparatide dose dependently improved cortical and trabecular microarchitecture of vertebra and femoral neck, as well as trabecular microarchitecture of the iliac crest. Bone microarchitecture at all sites was significantly correlated with lumbar vertebra and femoral neck strength. Cortical bone volume of vertebra had the strongest correlation with vertebral and femoral neck strength. Therefore, structural improvement seemed to be part of the mechanism for improved strength observed with teriparatide treatment. Trabecular bone architecture of the iliac crest at 6 months also correlated with vertebral and femoral neck strength, as did femoral neck (cortical and trabecular) histomorphometry and trabecular histomorphometry of vertebra after 18 months of treatment (6).

Table 2: Cortical bone volume (%) in lumbar vertebrae and femoral neck obtained after 18 months of teriparatide treatment in OVX monkeys (6)

Group	Lumbar vertebrae	Femoral neck
OVX (n = 20, control)	62.0 ± 10.8	91.3 ± 5.0
teriparatide (1 µg/kg per day, n = 19)	69.2 ± 8.8*	90.7 ± 3.3
teriparatide (5 µg/kg per day, n = 21)	72.1 ± 6.5*	94.3 ± 2.2*

OVX - ovariectomised monkeys; * $p < 0.05$ compared with OVX

Impact of age

Friedl et al. (2007) investigated whether aging impacts the bone anabolic response to teriparatide. Female Sprague-Dawley rats at 1, 3 and 13 months of age were either treated by teriparatide or by vehicle solution (control) for one week. Effects on static and dynamic histomorphometry of cancellous bone were the main outcome measures. There was a profound decrease in bone formation rate with aging in control rats, whereas active treatment resulted in a significant relative 1.5-, 3- and 4.7-fold increase in bone formation rate, without altering indices of bone resorption. Aging decreased and teriparatide increased mRNA levels for bone matrix proteins and growth factors in a gene-specific manner. In younger animals, teriparatide-induced a marked stimulation in the mineral apposition rate with no effect on osteoblast number, whereas the latter was increased in older animals (1.0-, 1.7- and 3.1-fold). Active treatment in young rats led to a significant increase in trabecular number (1.6 - 2.6/mm, $p < 0.05$), whereas older rats demonstrated increases in trabecular thickness only (52.8 - 77.8 µm, $p < 0.001$). Accordingly, although teriparatide increased bone formation at all ages, significant age-related differences in the cellular and molecular mechanisms involved in the bone anabolic response were noted (9).

Effect dose frequency

Watanabe et al. (2018) evaluated different dosing frequencies in Crl:CD(SD) rats (n = 60) to examine the varying levels of anabolic action of the drug. Sixty animals were treated with s.c. teriparatide over 3 months either once weekly or once daily. Within each group, three rats were assigned to the control group, and nine were assigned to each of the three dose groups. Once-a-week doses were 75, 100 or 125.4 µg/kg/week and once-a-day doses 5, 10 or 13.6 µg/kg/day. The levels of biomarkers in the blood were compared and found to vary in osteocalcin, a biomarker of bone formation, and cross-linked N-telopeptide of type 1 collagen (NTX), a biomarker of bone resorption, according to the dosing frequency. In the once-weekly regimen, teriparatide did not affect NTX levels at any of the doses studied, while osteocalcin levels increased with dose, peaking at 72 hours, then returning to normal before the next injection (after one week). BMD levels increased moderately with no difference between doses. This was thought to result from the equilibrium achieved following increases in bone formation and bone absorption. In the once-daily dosing regimen, meanwhile, NTX levels increased with dose, and osteocalcin levels were markedly higher when compared to those with the once-weekly dosing. BMD levels were higher than those with the once-weekly dosing, but with no difference between doses. This was considered a result of unlimited, excessive increases in bone formation due to daily administration of the drug. These results suggest that teriparatide promotes normal bone metabolism ("stationary

mini-modelling") when administered once weekly, and has an anabolic action with high metabolic turnover ("high-turnover remodelling") when administered once daily (28).

Sequential treatment with a bisphosphonate and teriparatide

Altman-Singles et al. (2017) aimed to determine the effect of intermittent teriparatide following 12 weeks of alendronate treatment on bone microarchitecture, bone remodelling dynamics and bone tissue material quality in 6-month-old OVX rats. While alendronate treatment effectively prevented additional bone loss, it also resulted in significant reductions in the heterogeneity of bone tissue mineral density and tissue modulus. Teriparatide treatment was able to shift the bone remodelling balance in favour of formation, with or without a prior alendronate treatment. Moreover, by altering the tissue mineralisation, teriparatide alleviated the reduction in heterogeneity of tissue material properties induced by prolonged alendronate treatment. Furthermore, switching to teriparatide treatment from alendronate improved bone's post-yield bone mechanical properties at both the whole bone and apparent level compared to alendronate alone (1).

Shimizu et al. (2017) evaluated sequential treatment with zoledronic acid and teriparatide (or vice versa) in OVX rats and assessed the effects on bone strength and bone turnover. Two months after ovariectomy, osteopenic rats (n = 172) were treated with zoledronic acid, teriparatide or vehicle for a period of 4 months (first treatment period), and then the treatments were switched and administered for another 4 months (second treatment period). The group treated with zoledronic acid followed by teriparatide showed an immediate increase in BMD of the proximal tibia and greater BMD and bone strength of the lumbar vertebral body, femoral diaphysis and proximal femur than the group treated with zoledronic acid followed by vehicle. Serum osteocalcin, as a marker of bone formation, increased rapidly after switching to teriparatide from zoledronic acid. The group treated with teriparatide followed by zoledronic acid did not lose bone mass in the proximal tibia after teriparatide was stopped, while the group treated with teriparatide followed by vehicle did lose bone mass. The BMD and bone strength of the lumbar vertebral body, femoral diaphysis and proximal femur were greater in the group treated with teriparatide followed by zoledronic acid than in the group treated with teriparatide followed by vehicle. The increase in serum osteocalcin and urinary type 1 collagen cross-linked C-telopeptide (CTX) after withdrawal of teriparatide was prevented by the switch from teriparatide to zoledronic acid. The results demonstrated that in osteopenic rats switching from zoledronic acid to teriparatide resulted in a non-attenuated anabolic response in the lumbar spine and femur of OVX rats. In addition, switching from teriparatide to zoledronic acid caused BMD to be maintained or further increased. The authors stated that in this animal model, follow-up treatment with zoledronic acid prevented BMD reductions and thus may be a suitable bisphosphonate for maintenance therapy of after discontinuation of teriparatide (21).

Combined use of anti-osteoporotic drugs

Zhang et al. (1998) examined the efficacy of concurrent treatment with teriparatide and bisphosphonate (incadronate) in augmenting cortical and cancellous bone mass of femoral neck in OVX rats. In OVX rats treated with teriparatide alone or teriparatide plus incadronate increased bone formation completely restored lost cancellous and cortical bone mass of femoral neck. The bone formation parameters and bone turnover seen with teriparatide plus incadronate were similar to those seen with teriparatide treatment alone, indicating that the bisphosphonate did not blunt the anabolic action of teriparatide when used concurrently (29).

Hodsman et al. (1999) tested the hypothesis that an antiresorptive agent might reduce the dosing requirement for an anabolic drug. In OVX rats, the concurrent use of raloxifene plus teriparatide

did not enhance the anabolic effects of the latter drug, but suggested that dual therapy may allow for reduced pharmacological doses of teriparatide needed to reverse oestrogen deficiency-induced osteopenia in rats (11).

Spinal fusion model

Both in rats and rabbits, teriparatide has been shown to enhance spinal fusion. To characterise a dose-dependent effect of teriparatide, Ming et al. (2012) took a trabecular bone graft which was implanted onto the L5 and L6 transverse processes of the same rat. Rats were randomly assigned into 3 groups: saline vehicle control, teriparatide 4 μg/kg per day (PTH4 group), and teriparatide 23 μg/kg per day (PTH23 group) s.c. injections for 4 weeks (5 day per week). The L5-L6 spinal segments were harvested at week 4, and assessments included radiography, micro-computed tomography, manual palpation and histomorphometry. The average radiographical score of L5-L6 fusion in vehicle, PTH4 and PTH23 groups was 1.53, 2.87, and 4.11, respectively, with the PTH23 being significantly higher ($p = 0.001$ vs. vehicle). The average micro-computed tomographic score of L5-L6 fusion in vehicle, PTH4 and PTH23 groups was 1.53, 2.40 and 3.74, respectively ($p = 0.001$, PTH23 vs. vehicle and PTH4). Manual palpation showed that fusion rate was 20%, 50% and 67.7% in vehicle, PTH4 and PTH23 groups, respectively. The bone mineralisation apposition rate at the fusion site was significantly increased in a dose-dependent manner among the groups. Teriparatide significantly increased vertebral and femoral BMD, bone mineral content and trabecular area in a dose-dependent manner relative to vehicle. No difference was found between the circulating procollagen type I N-terminal propeptide and intact osteocalcin levels in the serum at 4 weeks after treatments. Teriparatide at 23 μg/kg per day for 4 weeks showed anabolic skeletal effects and significantly enhanced spinal fusion rate in rats, whereas teriparatide at 4 μg/kg per day had also anabolic effects but did not significantly enhance spinal fusion rate (17).

Effects on osteonecrosis

Dong et al. (2015) evaluated the effect of s.c. injection of teriparatide for the steroid induced femoral head necrosis in a rat model. Twenty-four adult male Sprague-Dawley rats were randomised into 4 groups: 18 rats from LPS/MPS group, LPS/MPS+PTH group and LPS/MPS+NS group were given lipopolysaccharide (LPS, 20 μg/kg) and methylprednisolone (MPS, 40 mg/kg) to establish the steroid-induced osteonecrosis model. Six rats from NS group only received normal saline (NS). Four weeks later, all the rats in LPS/MPS group and NS group were sacrificed and the femoral heads were harvested. After that, the 6 rats in the LPS/MPS+PTH group received s.c. injection of 20 μg/kg teriparatide and LPS/MPS+NS group only received equal amount of normal saline. After 4 weeks, the serum bone marker was tested and the femoral heads were harvested. Micro-CT and histological examination were performed to compare the incidence of osteonecrosis and trabeculae parameters for the femoral head. At 4 weeks, rats in LPS/MPS group showed significant osteonecrosis by histological examination (83.3%) which suggested successful steroid induced osteonecrosis animal models were established. After the treatment of 4 weeks, the LPS/MPS+PTH group showed significant lower incidence rate of osteonecrosis compared with the LPS/MPS+NS group (16.7% vs. 75%, $p < 0.05$). The micro-CT examination showed a higher bone volume/total volume ratio, trabecula thickness and BMD in the LPS/MPS+PTH group compared with the LPS/MPS+NS group. The serum osteocalcin was a little higher in the LPS/MPS+PTH group (4.54 ± 1.61 vs. 3.58 ± 1.81, $p = 0.358$). Systemic application of teriparatide for steroid-induced osteonecrosis in rats showed a beneficial effect (8).

Promoting effects on bone healing / fracture repair

Fracture consolidation is a biological process resulting in the restoration of injured bone to a state of normal structure and function. Due to its anabolic properties, teriparatide has been investigated as a pharmacological stimulant. Midshaft closed fracture of the right tibia was manually performed

in a mouse model. To determine the most effective dose, mice were randomly subdivided into five groups and treated as follows: group 1 (n = 8), 4 µg/kg teriparatide daily; group 2 (n = 8), 20 µg/kg teriparatide daily; group 3 (n = 10), 40 µg/kg teriparatide daily; group 4 (n = 8), 40 µg/kg teriparatide every third day; control (n = 10), saline solution daily. Teriparatide's effect on callus formation was monitored during the first 4 weeks from fracture. Daily administration of 40 µg/kg accelerated callus mineralisation from day 9 onward without significant increase of sizes, and at day 15 the microhardness properties of treated callus were similar to those of bone tissue. The author stated that in this mouse model teriparatide considerably improved callus consolidation in the very early phases of bone healing (18).

To evaluate effects on fracture healing, 50 adult male Sprague-Dawley rats were subjected to a unilateral tibia fracture and received internal fixation with a Kirschner needle of once-weekly and once-daily s.c. injections of teriparatide (10 or 20 µg/kg). Four weeks later, the rats were euthanatised, and the fractured tibiae were assessed. Compared to the saline control group, either daily or weekly applied teriparatide significantly increased bone mass, improved the bone microarchitecture and promoted fracture healing ($p < 0.05$). There were no significant differences in BMD, bone microstructure or bone strength between the doses ($p > 0.05$) (30).

Since it has been reported that vitamin K‐dependent γ‐carboxylation of osteocalcin may enhance the efficacy of euproxine in bone repairing in OVX rats, Huang et al. (2018) tested the hypothesis that vitamin K combined with teriparatide can promote bone formation and inhibit bone degradation, thereby improving bone metabolism in a rat model of osteoporosis. Ovariectomised rats were subjected to oral intake of vitamin K or subcutaneous injection of teriparatide or both for 8 weeks. Enzyme-linked immunosorbent assays (ELISAs) were used to detect the content of carboxylated-type of osteocalcin (Gla-OC) and CTX in serum. Compared with monotherapy, vitamin K combined with teriparatide significantly increased serum Gla-OC level and the number of osteoblast, decreased serum CTX level, reduced the number of osteoclasts and increased bone density and strength. This study showed that the efficacy of vitamin K combined with teriparatide is better than that of monotherapy. However, further studies need to elucidate the molecular mechanism of the effects of vitamin K combined with teriparatide and their clinical relevance (12).

Grafting beta-tricalcium phosphate (TCP) is a well-established method for restoring bone defects; however, there is concern that the mechanical stability of the grafted beta-TCP is not maintained during bone translation. Komatsu et al. (2018) evaluated the effect of intermittent teriparatide treatment on changes in bone grafted with beta-TCP using a rabbit bone defect model. Bone defects (5 x 15 mm) were created in the distal femoral condyle of Japanese white rabbits, and beta-TCP granules of two different total porosities were manually grafted. Teriparatide (40 µg/kg) or 0.2% rabbit serum albumin solution as a vehicle control was s.c. injected three times per week following the surgery. At 4 or 8 weeks post-surgery, teriparatide treatment significantly increased ($p < 0.05$) the serum levels of Gla-OC, a specific marker for bone formation, suggesting that teriparatide enhances bone formation in beta-TCP-grafted rabbits. Furthermore, teriparatide increased the degradation of beta-TCP by bone remodelling ($p < 0.05$) and promoted the formation of new bone following application of the graft compared with the control group ($p < 0.01$). Furthermore, teriparatide suppressed the reduction in mechanical strength ($p < 0.05$) during bone translation in bone defects grafted with beta-TCP. The authors stated that, possibly by promoting new bone formation, teriparatide effectively maintained the mechanical stability of grafted beta-TCP (14).

In summary, the intermittent administration of teriparatide stimulated bone formation in various animal models including ovariectomised rats and monkeys. Beneficial effects were noted with respect to mass and strength of trabecular and cortical bone. Complementary positive actions were seen in case of concurrent and/or sequential application of bisphosphonates. In monkeys, positive bone changes persisted over months following the discontinuation of teriparatide. In experimental animal studies, teriparatide demonstrated promoting effects on bone healing and fracture repair.

2.4.2.3 Pharmacodynamic interactions

Concurrent administration of the selective oestrogen receptor modulator (SERM) raloxifene and teriparatide increased BMD in trabecular bone in osteopenic rats. Raloxifene did not modify the efficacy of teriparatide in rats (G2).

In rats, teriparatide alone or in combination with oestrogens, increased bone mass and bone strength in comparison with vehicle-treated rats. Administration of 1,25 dihydroxyvitamin D did not significantly affect the skeletal efficacy of teriparatide (G2).

Concomitant administration of teriparatide and antiresorptive agents neither blocked nor enhanced its skeletal effects on the femoral neck, femur, vertebrae or tibia of intact males or osteopenic female rats (G2).

In rats, growth hormone alone increased trabecular and cortical bone mass. A 30% to 50% greater bone mass was observed when growth hormone was combined with teriparatide. This synergetic effect disappeared in aged female rats (G2).

Findings from preclinical interactions studies suggested no potential for clinically relevant drug interactions.

2.4.2.4 Secondary and safety pharmacodynamics

Swami et al. (2017) hypothesised that intermittent teriparatide administration alters the bone microenvironment, rendering it less favourable for breast cancer cell colonisation. Using orthotopic and intratibial models of 4T1 murine and MDA-MB-231 human breast cancer tumours, the authors demonstrated that teriparatide decreases both tumour engraftment and the incidence of spontaneous skeletal metastasis in mice. Microcomputed tomography and histomorphometric analyses revealed that teriparatide increased bone volume and reduced tumour engraftment and volume. Transwell migration assays with murine and human breast cancer cells showed that teriparatide alters the gene expression profile of the metastatic niche, in particular vascular cell adhesion protein-1, to inhibit recruitment of cancer cells. While teriparatide did not affect growth or migration of the primary tumour, it elicited several changes in the tumour gene expression profile resulting in a less metastatic phenotype. As teriparatide had no promoting effect on the primary tumour, further evaluation in preventing breast cancer metastasis is warranted (22).

Decreased blood pressure and increased heart rate were observed in conscious rat and dog models reflecting teriparatide-induced vasodilation. The no-observed-effect level (NOEL) for cardiovascular changes in the rat was 4.3 µg/kg of teriparatide. In female dogs, a decrease of

arterial pressure and increase of left ventricular inotropic state and heart rate were observed after treatment with doses of 6 µg/kg per day (G2).

A quantitative assessment of electrocardiogram data after repeated doses of teriparatide did not show any effects on cardiac conduction, re-polarisation or production of cardiac arrhythmia (G2).

In male adult mice, teriparatide given at doses of 100 µg/kg did not produce secondary pharmacology effects related to the central nervous system and behavioural functions such as changes in body temperature (G2).

2.4.3 Pharmacokinetics

The scientific discussion on the originator product Forsteo (INN teriparatide, G2) summarised pharmacokinetic studies with single and repeated doses of teriparatide that were performed in Fischer 344 rats and cynomolgus monkeys.

Clearance and volume of distribution, measured after 10 µg/kg of teriparatide intravenous (i.v.) administration were 67.5 mL/min/kg and 0.54 L/kg in rat and 6.18 mL/min/kg and 0.14 L/kg in monkey, respectively. Minor sex differences were observed in rats with the higher parameters in males than in females (74.2 mL/min/kg and 62.1 mL/min/kg and 0.61 L/kg and 0.48 L/kg for clearance and volume of distribution, respectively) (G2).

The absolute bioavailability of the s.c. route for 10 µg/kg dose was 0.57 in rat (0.61 in males and 0.55 in females) and 0.36 in monkey (0.39 in males and 0.34 in females). The bioavailability of teriparatide was lower in animals than in humans (57% in rat and 36% in monkey for a 10 µg/kg dose versus 95% in humans) (G2).

The kinetic profile of teriparatide demonstrates a short t_{max} and elimination half-life (both 15 -40 min in the rat and monkey) after s.c. injection. The area under the concentration time curve (AUC) and maximum concentration (C_{max}) were dose-related. Minor differences in C_{max} and AUC were observed between different toxicokinetic studies in rats, but no consistent pattern of increase or decrease and no consistent effect of gender could be noted. There were no changes in terminal elimination rates after repeated dosing; no serum accumulation or enzyme induction was detected following repeated administration. No notable gender differences were observed in both species, except in rats at high teriparatide doses (i.e. > 100 µg/kg for C_{max} and AUC values greater in female than in male) (G2).

In male and female cynomolgus monkeys given daily s.c. doses of teriparatide for 3 months, no induction of hepatic microsomal enzymes was noted (G2).

Serada et al. (2012) characterised the pharmacokinetics of teriparatide acetate in rats after s.c. administration. Teriparatide was rapidly absorbed into the circulation and eliminated immediately. No intact teriparatide was detected in the urine. To elucidate the mechanism of teriparatide metabolism, both *in vivo* and *in vitro* studies were performed using a radiolabelled (^{125}I)-teriparatide analogue. Following s.c. administration, the concentration of analogue metabolites increased in the plasma time-dependently. The concentration in the kidneys was more than 3-fold the concentration in the liver. *In vitro* analyses, in which the ^{125}I-labelled analogue was added to blood, liver and kidney tissues obtained from male rats, suggested that kidney radioactivity was associated with the degraded bioactive analogue. In model rats with renal and hepatic dysfunction,

renal failure but not hepatic failure affected the pharmacokinetics of teriparatide, which accounted for the decrease in the clearance of teriparatide. The authors concluded that in rats, s.c. administered teriparatide is rapidly absorbed and distributed to the liver or kidneys, where it is immediately degraded (20).

Drug interactions
Relevant information regarding preclinical pharmacokinetic drug interaction studies was not detected in the literature.

2.4.4 Toxicology

2.4.4.1 Single dose toxicity

Acute toxicity studies in rats using s.c. (doses up to 1000 µg/kg) or i.v. (doses up to 300 µg/kg) administration of teriparatide did not reveal any functional toxic effects and the observable effects seemed to be related to the vasodilatation effects of the active substance (G2).

2.4.4.2 Repeated dose toxicity

Repeated dose toxicity studies were conducted in rats for up to 6 months using doses up to 300 µg/kg/day and in cynomolgus monkeys for up to one year using doses up to 10 µg/kg/day with s.c. administration (G2, G5).

In cynomolgus monkeys, two repeated dose toxicity studies have been carried-out, one study evaluated effects on renal function (4 months of treatment with 3-months reversibility period) and the other evaluated histopathologic patterns (8 months of treatment or 12 months of treatment with 6-month reversibility period). Histological changes observed in the renal function study are probably related to an exaggerated pharmacological effect induced by hypercalcaemia (G2, G5).

The scientific discussion on the originator product Forsteo (INN teriparatide) published in 2005 concluded that toxicity studies revealed a similar pattern of findings, which resulted from exaggerated pharmacological effects of high doses of teriparatide. The main target organs were the bone, liver spleen and kidney. Data suggest that the renal changes observed are secondary to increased calcium mobilisation. Since hypercalcaemia is not observed in patients at the intended clinical dose, the occurrence of kidney lesions seems unlikely. The changes are more evident in rats than in monkeys (G2).

As regards the immunogenic potential of teriparatide, an absence of antigenic response was reported in rodents and a weak antigenic potential in monkeys (G2).

Results reported from a long-term oncogenicity study in rats treated with daily teriparatide suggested a drug- and dose-related incidence of osteosarcoma (see section 2.4.4.4) (3) (G5).

2.4.4.3 Genotoxicity and immunotoxicity

Teriparatide was not genotoxic in *in vitro* and *in vivo* genotoxicity tests, including the *in vitro* mutagenesis assay with and without activation, mouse lymphoma assay for mammalian cell

mutation, the Chinese hamster ovary chromosomal aberration assay, and the *in vivo* micronucleus test in the mouse (5) (G2, G5).

2.4.4.4 Carcinogenicity

In an initial oncogenicity study, Fischer 344 (F344) female and male rats, approx. 6 to 8 weeks of age, were administered 0, 5, 30 or 75 µg/kg/day teriparatide for 2 years. Exaggerated bone formation and formation of osteosarcomas were noted in all treatment groups, raising concern regarding the potential development of bone tumours in humans treated with teriparatide (23, 25).

As the initial study findings strongly suggested that the administered dose as well as the duration of treatment were important factors in the development of bone neoplasms, Vahle et al. (2004) conducted an additional long-term study in female F344 rats to determine the relative importance of dose, treatment duration and age at initiation of treatment on the incidence of teriparatide-induced bone proliferative lesions. Treatment groups consisted of different combinations of dose (0, 5 or 30 µg/kg/day), treatment duration (6, 20 or 24 months) and age at initiation of treatment (2 or 6 months of age). The primary endpoints were the incidence of bone neoplasms and effects on bone mass and structure as evaluated by quantitative computed tomography (QCT) and histomorphometery. Significant increases in the incidence of bone tumours (osteoma, osteoblastoma and osteosarcoma) occurred in rats treated with 30 µg/kg/day for 20 or 24 months. No neoplasms were found when the 5 µg/kg/day treatment was initiated at 6 months of age and continued for either 6 or 20 months (up to 70% of lifespan). This treatment regimen defined a "no-effect" dose for neoplasm formation that nevertheless resulted in substantial increases in bone mass. According to the authors, treatment duration and administered dose are the most important factors in the teriparatide-induced bone tumours in rats (25).

Subsequent studies in F344 rats revealed a non-carcinogenic dose level of 10 µg/kg/day with 2-year daily s.c. administration. Watanabe et al. (2012) reported that for male and female Sprague-Dawley rats treated with this regimen, the incidence of osteosarcoma was increased at 13.6 µg/kg/day. The non-carcinogenic dose level was 4.5 µg/kg/day both for male and female animals. Responses of the bones to teriparatide were similar between F344 and Sprague-Dawley rats in many aspects. These results suggested that the carcinogenic potential of teriparatide in Sprague-Dawley rats is essentially the same as in F344 rats (27).

Bone tumours were not observed in studies in OVX cynomolgus monkeys, but these studies were not prospectively designed to detect bone tumours and did not include a prolonged post-treatment observation period to determine whether neoplasms might arise after cessation of teriparatide treatment. Therefore, Vahle et al. (2008) collected prospective data in OVX, skeletally mature cynomolgus monkeys (n = 30 per group) that were given teriparatide for 18 months at either 0 or 5 µg/kg/day s.c. After 18 months of treatment, subgroups of six monkeys from both groups were killed and evaluated, whereas all remaining monkeys entered a 3-year observation period in which they did not receive teriparatide. Surveillance for bone tumours was accomplished with plain film radiographs, visual examination of the skeleton at necropsy and histologic evaluation of multiple skeletal sites. After the 18-month treatment period, vertebral BMD, bone mineral content (BMC) and strength (ultimate load) were increased by 29%, 36% and 52%, respectively, compared with controls. Proximal femur BMD, BMC and strength were also increased by 15%, 28% and 33%, respectively. After 3 years without treatment, no differences in bone mass or strength at the vertebra were observed relative to controls; however, the femoral neck showed significant persistence in stiffness (20%), BMC (14%) and trabecular bone volume (53%), indicating a

retention of teriparatide efficacy at the hip. Radiographs and histology did not identify any bone proliferative lesions or microscopic lesions of osteosarcoma at the end of the treatment or observation period. These data indicate that teriparatide did not induce bone proliferative lesions over a 4.5-year interval of observation, but the small group size limit the ability to draw definitive conclusions regarding the risk of bone tumour developments in patients (26).

While relevance of the observations regarding osteosarcoma in rats for humans cannot be completely ruled out, the following should be noted: Rats were treated daily for up to 24 months, which is 80 - 90% of the normal rat life span. The duration of therapy in humans is approx. 18 - 24 months, about 2% of the human life span. Rats were treated for 25 - 30 bone-turnover cycles, while humans will only be treated for approximately 1 - 3 bone-turnover cycles. In addition, there are fundamental differences between rat and primate / human bone physiology and subsequent responses to teriparatide which is underlined by the observations made in monkeys (5).

Given that rats and mice are bone-modelling species (i.e. bone resorption and formation occur at different sites) but rabbits, monkeys and humans are bone-remodelling species (i.e. the bone undergoes a continuous, coordinated process of bone resorption followed by new bone formation at the same site), the relevance to humans of the findings in the carcinogenicity studies is unlikely. This assessment is supported by human data collected over 15 years. However, as osteosarcoma formation in rats was clearly dependent on treatment duration, and without fully understanding the mechanism of tumour formation, the clinical treatment duration is restricted to 24 months and osteosarcoma is a potential risk for humans.

2.4.4.5 Reproductive and developmental toxicity

In pregnant rats given subcutaneous teriparatide doses up to 1000 µg/kg/day, there were no findings. In pregnant mice given subcutaneous doses of 225 or 1000 µg/kg/day from gestation day 6 through 15, the foetuses showed an increased incidence of skeletal deviations or variations (interrupted rib, extra vertebra or rib) (G5).

Developmental effects in a perinatal/postnatal study in pregnant rats given s.c. doses of teriparatide from gestation day 6 through postpartum day 20 included mild growth retardation in female offspring at doses ≥ 225 µg/kg/day (≥ 120 times the human dose based on surface area), and in male offspring at 1000 µg/kg/day. There was also reduced motor activity in both male and female offspring at 1000 µg/kg/day. There were no developmental or reproductive effects in mice or rats at a dose of 30 µg/kg/day (G5).

No effects on fertility were observed in male and female rats given subcutaneous teriparatide doses of 30, 100, or 300 µg/kg/day prior to mating and in females continuing through gestation day 6 (G5).

Reproductive toxicity studies were generally uneventful indicating a low potential for effects on male and female fertility and embryo toxicity. However, there was some evidence for growth retardation and reduced motor activity in F1 animals in the rat pre- and postnatal study. In addition, teriparatide was associated with a high level of embryo lethality in the rabbit, probably associated with the foetal sensitivity of this species to hypercalcaemia. Given these uncertainties, it is considered prudent to avoid foetal exposure to teriparatide (G2).

2.4.4.6 Local tolerance

Non-clinical studies using the subcutaneous route have not been conducted to specifically investigate local tolerance of teriparatide. However, studies in animals using this route did not reveal any significant irritation at the injection site. Mild erythema and/or irritation were related to the procedure itself and not to teriparatide (G5).

2.4.4.7 Environmental risk assessment

Teriparatide is a recombinant human peptide with a comparatively simple structure (no posttranslational modifications) and therefore not expected to pose a risk to the environment.

2.4.5 Excipients

The choice of excipients is based on experience and compatibility of the chosen excipients with the active substance. All excipients comply with their respective European Pharmacopoeia monograph. The excipients used for the product under discussion are well known, accepted and commonly used ingredients for the formulation of solutions for injection or infusion with most of them being listed in the Food and Drug Administration (FDA) database for inactive ingredients in approved drug products (G8). They will not modify the safety profile of the medicinal product under discussion. Therefore, none of these inactive ingredients need any special comment and can be assigned as appropriate for the intended purpose.

Acetic acid, glacial
Glacial and diluted acetic acid solutions are widely used as acidifying agents in a variety of pharmaceutical formulations and food preparations. In pharmaceutical products, acetic acid is widely used as a buffer system when combined with an acetate salt such as sodium acetate. Acetic acid is also claimed to have some antibacterial and antifungal properties. The diluted acid is generally regarded as relatively nontoxic and non-irritant (G7). The FDA lists acetic acid as a generally recognised as safe (GRAS) substance (G9). As an acidifying agent, it is accepted for use as a food additive in Europe (G10). No adverse effects are expected with respect to the product under review, which contains 0.41 mg glacial acetic acid per mL solution.

Sodium acetate
Acetate is the conjugate base of acetic acid and the salt sodium acetate is commonly used in pharmaceutical products as component of a buffer system. Sodium acetate is contained in the FDA database for inactive ingredients in approved drug products with various routes of administration including subcutaneous use (G8). No adverse effects are expected with respect to the product under review, which contains 0.10 mg sodium acetate per mL solution.

Mannitol
Mannitol is a hexahydric alcohol related to mannose and isomeric with sorbitol. It is widely used in pharmaceutical formulations and food products, for example as a tablet diluent or excipient for chewable tablets. In parenteral formulations, mannitol functions as a tonicity adjusting agent (G7). It is contained in the FDA database for inactive ingredients in approved drug products with various routes of administration including subcutaneous application (G8) and is classified as GRAS by the

FDA (G9). No adverse effects are expected with respect to the product under review, which contains 45.4 mg mannitol per mL solution.

Metacresol
Metacresol (m-cresol) or methylphenol is commonly used in pharmaceutical products for its antimicrobial activity. Reports of adverse reactions to metacresol are generally associated with the use of either the bulk material or cresol-based disinfectants, which may contain up to 50% metacresol rather than for its use as a preservative. Only isolated cutaneous hypersensitivity reactions to metacresol have been reported (G7). Metacresol is contained in the FDA database for inactive ingredients in approved drug products (G8). No adverse effects are expected with respect to the product under review, which contains 3.0 mg metacresol per mL solution.

Hydrochloric acid
Hydrochloric acid is widely used as an acidifying agent, in a variety of pharmaceutical and food preparations. Whereas the concentrated solution is corrosive and can cause severe damage on contact with the eyes and skin, when used diluted, at low concentration, hydrochloric acid is not usually associated with any adverse effects. The compound is accepted for use as a food additive in Europe (G10). The FDA lists hydrochloric acid as a GRAS substance (G9) and it is also included in the FDA database for inactive ingredients in approved drug products (G8). Thus, no adverse effects are expected with respect to the product under review, in which hydrochloric acid is used for pH adjustment.

Sodium hydroxide
Sodium hydroxide is widely used in pharmaceutical formulations to adjust the pH of solutions. It can also be used to react with weak acids to form salts. Sodium hydroxide is widely used in the pharmaceutical and food industries and is generally regarded as a nontoxic material at low concentrations. At high concentrations, however, it is a corrosive irritant to the skin, eyes and mucous membranes (G7). The FDA lists sodium hydroxide as a GRAS substance (G9) and it is also included in the FDA database for inactive ingredients in approved drug products (G8). Thus, no adverse effects are expected with respect to the product under review, in which sodium hydroxide is used for pH adjustment.

Water (for injection)
Water is widely used as a raw material, ingredient and solvent in the processing, formulation and manufacture of pharmaceutical products. Water for pharmaceutical purposes is prepared by distillation, by ion exchange, by reverse osmosis or by any other suitable method that complies with the regulations on water intended for parenteral use in humans. The Ph. Eur. states that water for injections is produced by distillation (G7). No adverse effects are expected with respect to the product under review, which contains water for injections as a solvent.

In conclusion, the excipients contained in the product TERIPARATIDE are of the appropriate pharmaceutical grade, are assigned as appropriate and safe by pertinent databases and thus do not pose any hazard to the intended patient population.

2.4.6 Integrated overview and conclusions

The medicinal product TERIPARATIDE 20 micrograms/80 microliters solution for injection in pre-filled pen contains teriparatide which is a 34-amino acid peptide identical in sequence to the N-terminal fragment (the biologically active region) of the naturally secreted 84-amino acid human parathyroid hormone (PTH).

Teriparatide can be either manufactured by chemical solid phase peptide synthesis (as in the product under discussion) or produced in *Escherichia coli* using recombinant DNA technology (as in the originator / reference product Forsteo). Of note, the preclinical and clinical characteristics of teriparatide are independent of its mode of synthesis.

Teriparatide's mechanisms of action are comparable to those of endogenous PTH. In response to a decrease in plasma calcium ions, PTH restores calcium levels to normal via a number of mechanisms: release of calcium from the bone, reabsorption of calcium by the kidney and, indirectly, renal synthesis of 1,25-dihydroxyvitamin D, which subsequently increases the absorption of intestinal calcium. The endogenous hormone also regulates reabsorption of phosphate by the kidneys. Teriparatide and the 34 N-terminal amino acids of PTH bind to the same receptors with the same affinity and thus produce the same physiological effects on bone and kidneys.

The pattern of systemic exposure determines the skeletal response to teriparatide. While continuous availability favours bone resorption, intermittent exposure stimulates bone formation. In rat and monkey models, once-daily subcutaneous administration over several months enhanced bone mass and improved bone microarchitecture and mechanical strength. Accompanying changes in bone markers indicating increased bone formation were also documented. Several studies in rats demonstrated beneficial effects when teriparatide and bisphosphonates were administered concurrently and/or sequentially. In monkeys which had been treated with teriparatide over 18 months, improved skeletal mass was sustained at evaluation 6 months after drug discontinuation. A number of experimental studies reported promoting effects of teriparatide on bone healing and/or fracture repair.

The pharmacokinetic profile of teriparatide was investigated in rats and monkeys following subcutaneous administration. Time to maximum concentration and elimination half-life were short in both species (i.e. 15 - 40 min). Bioavailability was lower in these animals (57% and 36%, respectively) compared to humans (~95%). Single versus repeated dosing did not significantly change these parameters; there were no signs of enzyme induction or accumulation. Although limited data were collected, preclinical evaluations did not indicate a potential for clinically relevant drug interactions with teriparatide.

In all studies, teriparatide administration was well tolerated and no acute toxicity was detected with single or repeated doses. In rats, mild cardiovascular effects were noted in association with vasodilation induced by teriparatide. In monkeys, minor effects on the kidneys were noted in functional and histological analyses; these were attributed to transient hypercalcaemia. A significant immunogenic potential was not observed in animal studies.

Teriparatide was not genotoxic in *in vitro* and *in vivo* genotoxicity tests. In rats treated over a significant lifespan teriparatide caused a dose-dependent increase in the incidences of both benign and malignant bone tumours. The effect was observed at systemic exposures ranging from 3 to 60 times the exposure in humans given a 20-µg dose of teriparatide. The carcinogenic potential of

teriparatide in rodents was attributed to the mode of bone formation and bone remodelling in these animals which is different in primates. In line with this assessment, no tumours were observed when teriparatide was given to cynomolgus monkeys for prolonged periods. Hence, observations in rat studies may not be predictive of the human risk for bone neoplasms. However, as a risk cannot be completely ruled out, osteosarcoma is classified as an important potential risk to patients treated with teriparatide and maximum lifetime exposure is limited to 24 months. In animal studies, teriparatide's potential for reproductive and embryo toxicity was overall low. Local tolerance with subcutaneous administration was very good.

As regards the excipients contained in the final product, they are all well-known, accepted and commonly used ingredients for the formulation of solutions for subcutaneous injection.

Teriparatide was introduced in the pharmaceutical market in the early 2000s. The reference product Forsteo / Forteo (INN teriparatide) became first available in the year 2002 in the United States and in 2003 in the European Union. Currently, teriparatide is authorised in numerous countries worldwide and its use in the treatment of osteoporosis of various origin is well established. From more than 15 years of clinical experience with medicinal products containing teriparatide, extensive data has been obtained on the incidence and frequency of adverse events in association with the use of the peptide.

Hence, the use of teriparatide in the treatment of osteoporosis and prevention of fragility fractures is well documented. The characteristics of teriparatide in the treatment of osteoporosis in postmenopausal women and in osteoporotic in men as well as in sustained systemic glucocorticoid therapy in women and men at high risk for fracture are adequately reflected in the proposed SmPC, and the indications and precautions for use are justified by the pharmacological and toxicological properties of teriparatide. It is therefore recommended to grant marketing authorisation for TERIPARATIDE 20 micrograms/80 microliters solution for injection in pre-filled pen under the conditions specified in the proposed SmPC.

2.4.7 References

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Acetic Acid, Glacial (page 5 - 6)

Mannitol (page 424 - 428)

Metacresol (Cresol, page 203 - 205)

Sodium hydroxide (page 648 - 649)

Hydrochloric acid (page 308 - 309)

Water (page 766 - 770)

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