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Novel mechanisms of action of Glucagon-like peptide-1 (GLP-1) receptor agonists on the skeleton

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Short title: GLP-1 agonists and bone

Abstract

Type 2 Diabetes Mellitus (T2DM) leads to bone fragility and predisposes to increased risk of fracture, poor bone healing and other skeletal complications. In addition, some anti-diabetic therapies for T2DM can have notable detrimental skeletal effects. Thus an appropriate therapeutic strategy for T2DM should not only be effective in re-establishing good glycaemic control but also in minimising skeletal complications. There is increasing evidence that Glucagon-like peptide-1 receptor (GLP-1r) agonists, now greatly prescribed for the treatment of T2DM, have beneficial skeletal effects although the underlying mechanisms are not completely understood. This review provides an overview of the direct and indirect effects of GLP-1RAs on bone physiology, focusing on bone quality and novel mechanisms of action on the vasculature and hormonal regulation. The overall experimental studies indicate significant positive skeletal effects of GLP-1RAs on bone quality and strength although their mechanisms of actions may differ according to various GLP-1RAs and clinical studies supporting their bone protective effects are still lacking. The possibility that GLP-1RAs could improve blood supply to bone, which is essential for skeletal health, is of major interest and suggests that GLP-1 anti-diabetic therapy could benefit the rising number of elderly T2DM patients with osteoporosis and high fracture risk.

Lay Abstract

Bone weakening is an important complication in individuals with type 2 diabetes (T2DM). This review summarises the effects on skeletal health of drugs that are similar to the hormone Glucagon-like peptide-1 (GLP-1), which are now used increasingly for the treatment of T2DM and could lead to a stronger skeleton.

1 Introduction

Diabetes mellitus (DM) is a chronic disease that progresses worldwide at alarming rates. For instance, in 2013, it has been estimated that DM affected 382 million individuals [1]. Projections for 2035 indicate a global burden of 55% to reach up to 592 million individuals [1]. Associated complications are commonly cardio-vascular events, nephropathy, retinopathy, neuropathy and bone fragility that dampen the quality of life of affected individuals.

Type 2 diabetes mellitus (T2DM) is by far the most common form of DM and is characterised by chronic hyperglycaemia and hyperinsulinemia mostly caused by insulin resistance (IR) in peripheral tissues such as the liver and muscle. The aetiology of bone fragility in T2DM is unclear. Indeed, bone mineral density is normal or slightly elevated in T2DM despite an increase risk of femoral neck fracture, suggesting alterations of bone "quality" rather than bone mass [2-4]. Bone quality is an umbrella term that regroups factors such as bone microarchitectures, tissue material properties and bone toughness [5]. Another important contributor for bone fracture is represented by an increased risk in falls in this population [6, 7]. At the cellular and molecular levels, T2DM is characterised by a reduction in bone turnover suggesting modifications of bone cell behaviours [8]. Furthermore, low testosterone and vitamin D levels, and high plasma sclerostin, are common features observed in T2DM patients [9].

Current treatment options of T2DM rely on lifestyle intervention and oral or injectable drugs, when needed, to reach an HbA_{1C} level of 7% or less. Among the most prescribed drugs, the glucagon-like peptide-1 receptor agonists (GLP-1RAs) have recently attracted attention as *Glp-1r* knockout animals and GLP-1 supplemented animals exhibited modifications of bone strength and quality as described below.

Endogenously, GLP-1 is produced by post-translational processing of the glucagon gene in enteroendocrine cells, mainly L-cells [10]. Two forms of GLP-1 are produced in the intestine, GLP-$1_{7\text{-}36NH2}$ and GLP-$1_{7\text{-}37}$ although the major circulating form is GLP-$1_{7\text{-}36NH2}$ [11]. L-cells are an open type endocrine cells highly polarised with secretory granules at their basolateral pole ready to be released in the capillary network running through the *lamina propria*. This secretion is regulated by intraluminal contents, neural stimuli and hormones [12]. Beyond its endocrine mode of action, GLP-1 has also been suspected to act via the autonomous nervous system and hypothalamic and brainstem nuclei [13].

To act, GLP-1 engages its receptor; the GLP-1r that is coded by the human *GLP1R* gene comprising 13 exons that span approximately 13.8kb [14] and localised on chromosome 6p21 [15]. The GLP-1r is expressed in the endocrine pancreas, gastro-intestinal tract, lung, heart, kidney and several regions of the brain [12]. Recent evidences also suggest that GLP-1 can bind in specific circumstances to the glucagon receptor [16]. The principal physiological role of GLP-1 is to potentiate glucose-dependent insulin secretion [17]. Extrapancreatic actions of GLP-1 results in reduction of food intake through the CNS, inhibition of gastric emptying, positive actions on the cardiovascular system and a role in energy expenditure [17].

GLP-1RAs are GLP-1 with extended half-life to be more resistant to degradation by the dipeptidyl peptidase-4 (DPP-4) enzyme. Several molecules listed in Table 1 have been developed by the pharmaceutical industry and now been approved for the treatment of T2DM. The aim of the present review is to provide the reader with a comprehensive analysis of the effects of GLP-1RAs on bone physiology with special focuses on the mode of action including effects on bone quality, blood flow to bone, and on the hormonal regulation of bone metabolism.

2 Skeletal effects of GLP-1 agonists: direct and/or indirect mechanisms of action

2.1 Clinical studies

Clinical data on the skeletal effects of GLP-1RAs are scarce. Bone turnover markers and bone mineral density have been assessed in T2DM patients treated with exenatide and liraglutide. However, all these studies reported no effects of GLP-1RA treatment on circulating bone markers or bone mineral density [18-20]. Interestingly, the effects of liraglutide administration on bone turnover markers have been reported not in diabetic but in the obese population for the weight-loss action of liraglutide. In that study, bone formation was improved as indicated by higher values for N-terminal propeptide of type 1 procollagen reported in the liraglutide arm, but no effects on bone resorption were observed [21].

Two meta-analyses have also been performed on the use of GLP-1RA and the possible effects of these medications on fracture risk. One meta-analysis found beneficial effects of liraglutide in reducing the occurrence of bone fractures [22]. The other meta-analysis found

neutral effect of GLP-1RA as compared with other anti-diabetic medications [23]. Interestingly, Driessen and colleagues [24, 25] investigated in the British and Danish populations the incidence of bone fracture in GLP-1RA takers as compared with non-takers. No significant difference was observed and they suggested that the effect of GLP-1RAs was neutral in the human diabetic population.

2.2 *In vivo* studies

The first understanding of GLP-1 actions in skeletal physiology arises from *Glp1-r* KO mouse. At 10 weeks of age, these mice exhibited a small reduction in bone mass associated with an increased number of osteoclasts and eroded surfaces [26]. On the other hand, the mineral apposition and bone formation rates appeared unaffected by GLP-1r inactivation [26]. Similarly, observations in the same KO model at 16 weeks of age and in the double incretin receptor knockout model at 26 weeks of age corroborated these findings [27, 28]. Taken together these results suggested a control of bone resorption (osteoclast differentiation and/or action) by the GLP-1r. According to the literature, this effect on resorption seems to be indirect through a reduction in calcitonin gene expression in GLP-1r-deficient animals [26] but further evidences are warranted.

While it is well established that GLP-1RAs increase bone mass (see paragraph 3), previous investigations of their effects on bone turnover in rodents are conflicting. It has been reported that 3 µg/kg/day and 4.2 µg/kg/day exenatide induced bone formation by osteoblast activation in old ovariectomised (OVX) rats [29] and in hindlimb-unloading rats [30] by promoting the osteogenic differentiation and inhibiting BMSC adipogenic differentiation. A decrease of osteoclastic surfaces was also observed [29]. In contrast, we found no effect of both 10 µg/kg/day exenatide and 0.3 mg/kg/day liraglutide on bone formation in OVX mice and a slight increase of osteoclastic surfaces with the drug [31]. Moreover, our recent results suggest that exenatide increases bone formation in a T2DM mouse model but not in control mice while there was no effect of the drug on bone resorption (Pereira M, Gohin S, Roux JP, Cleasby ME, Mabilleau G, Chenu C, manuscript in revision). Thus, it is still unclear whether the mechanism of action of GLP-1RAs in bone is direct or indirect and targeting bone formation or resorption.

2.3 *In vitro* studies

While several studies have reported that GLP-1RAs could have beneficial effects on the skeleton, the downstream molecular mechanisms underlying the osteogenic effect have not been identified [32, 33]. It is indeed unclear whether the mechanism of action of GLP-1RAs in bone is direct, through a functional GLP-1r expressed by bone cells, or indirect, via an increase in calcitonin production by the thyroid C-cell which inhibits bone resorption [26]. Furthermore, the presence and the identity of the GLP-1r in bone were controversial until recently and thus the basis for direct skeletal effects of GLP-1 has not been established.

We recently demonstrated that GLP-1 might directly affect bone cells via a GLP-1r identified in primary mouse osteoblasts isolated from calvaria and bone marrow-derived osteoclasts [31] and this was confirmed *in situ* using a GLP-1r antibody (abcam). Similarly, other studies showed that mouse osteoblast-like MC3T3-E1 cells express a functional receptor for GLP-1 [34]. However this receptor could be different from the cAMP-linked GLP-1r expressed in the pancreas, but its existence implies that a direct skeletal action of GLP-1 may be possible [34, 35]. In contrast, expression of the pancreatic-type GLP-1r mRNA was identified in human osteoblastic cell lines deriving from osteosarcomas, but its expression was dependent on the stage of osteoblastic development [36]. However, other study failed to demonstrate the presence of GLP-1r at the mRNA level in primary murine osteoblasts or osteoclasts [37]. Similarly, the presence of the pancreatic GLP-1r in osteocytic cells was controversial as it has been reported in some cell lines, but not all [31, 38], as well as in osteocytes in rat femurs [38].

The presence of GLP-1r in bone cells *in vitro* and *in situ* implies that GLP-1RAs could have direct effects on bone cells. A study has indeed identified potential skeletal beneficial effects of 10 nM of exenatide by promoting osteoblastogenesis and restraining adipogenesis through a β-catenin pathway, during BMMSC differentiation [30]. Despite increased osteoblastogenesis, no direct effect of GLP-1RAs on bone nodule mineralisation *in vitro* was shown with up to 100uM of exenatide and 1000nM of liraglutide [29, 31]. It is well established that exposure of primary osteoblast cells to high glucose levels inhibits *in vitro* bone nodule formation [39, 40]. Interestingly, despite no effect of exenatide on bone formation in normal glucose conditions, our results demonstrate that it can reduce the deleterious effect of glucose on bone formation *in vitro*, in a dose-dependent manner (Pereira M, Gohin S, Roux JP, Cleasby ME, Mabilleau G, Chenu C, manuscript in revision). This could be due to upregulated GLP-1r expression in high glucose conditions, which could in turn magnify the effect of GLP-1RAs [34]. Regarding the effects of GLP-1RAs on

osteoclastogenesis *in vitro*, we showed that both liraglutide and exenatide increased osteoclastogenesis, while decreasing the area resorbed per osteoclast, suggesting that GLP-1RAs stimulate osteoclastic differentiation but impair their resorptive activity [31].

3 GLP-1RAs and bone quality

Unfortunately, as neither peripheral quantitative computed tomography (pQCT) nor iliac crest bone biopsy are part of the usual care in diabetic clinical trials, human data on the effects of GLP-1RAs on all aspects of bone quality are presently missing. As such, the following summary of action of GLP-1RA is based on pre-clinical data obtained in animal models. Several animal models of either osteoporosis or T2DM have been used to assess the effects of two GLP-1RAs, exenatide and liraglutide, on bone quality and strength. However, data concerning potential bone effects of other GLP-1RAs, and especially those administered once weekly, are currently missing. Mice presenting a deletion of GLP-1r have also been generated and represented a suitable model to investigate the role of the GLP-1/GLP-r pathway in bone.

3.1 Effects of GLP-1RAs on bone strength

Our knowledge of the effects of the GLP-1/GLP-1r pathway on bone strength has been markedly improved by the use of *Glp-1r* KO mice. Indeed, although these animals are not diabetic, they exhibited a significant reduction in bone strength represented by lower ultimate load and stiffness [37]. Bone strength in response to GLP-1RA has also been investigated in osteoporotic animal models generated either by ovariectomy or disuse. In ovariectomy-induced osteoporosis, the use of exenatide at a concentration as low as 1 µg/kg/day for 16 weeks, led to improvement in maximum load and stiffness as well as Young's modulus and ultimate stress, suggesting amelioration in bone microarchitecture and/or tissue material properties [29]. In the rat tail suspension model, the administration of exenatide (4.2 µg/kg/day) for 4 weeks resulted in higher value for maximum loading, stiffness, stress and Young's modulus, suggesting here again ameliorations in bone microarchitecture and/or tissue material properties [30]. Our very recent data (Pereira M, Gohin S, Roux JP, Cleasby ME, Mabilleau G, Chenu C, manuscript in revision) showed a reduction in bone strength of diabetic db/db mice with significantly lower values for ultimate load, stiffness and work-to-failure, but four weeks administration of exenatide in these diabetic animals did not have any significant effect on bone strength parameters.

3.2 Effects of GLP-1RAs on bone microarchitecture

In *Glp-1r* KO animals, unpublished observations from our group, revealed that these animals presented with a reduction in cancellous bone volume associated with a lower trabeculae numbers and higher trabecular spacing. These data have been confirmed by the elegant study of Yamada et al. [26] who reported a significant reduction in cancellous bone mineral density in the same transgenic animal model. Alterations of cortical bone in this mouse model were also evidenced with lower outer bone diameter and cortical thickness [37]. Exenatide and liraglutide have been used as a treatment option in pre-clinical animal models of osteoporosis. Exenatide demonstrated positive effects on trabecular bone microarchitecture in the axial and appendicular skeleton evidenced by amelioration of structural parameters in lumbar vertebra and long bones as early as 4 weeks treatment. Indeed, exenatide resulted in higher bone volume/total volume (BV/TV) values (24% to 148%, depending on dose and treatment duration) and higher values for trabecular number (Tb.N), thickness (Tb.Th) and reduction in separation (Tb.Sp) [29-31, 41]. Liraglutide also showed improvement in trabecular bone microarchitecture but these effects seemed restricted to long bones with very poor action on the axial skeleton [31, 42]. The effects of GLP-1RA on cortical microarchitecture were only observed after a minimum of 8 weeks treatment with exenatide or liraglutide, but highlighted significant increases in cortical thickness with 20µg/kg/day of exenatide or 0.6mg/kg/day of liraglutide [41, 42].

In opposition to what is commonly observed in humans, animal models of T2DM exhibit significant alteration of trabecular and cortical microarchitectures. The effects of GLP-1RA use in diabetic animal models have also been reported. The use of exenatide at a regimen of 10 µg/kg/day for 3 days in T2DM animals resulted in improvement in trabecular microarchitecture at the femur and in the lumbar spine [43, 44]. Four weeks treatment with 10 µg/kg/day of exenatide also improved trabecular bone mass and architecture in the tibiae of db/db mice (Pereira M, Gohin S, Roux JP, Cleasby ME, Mabilleau G, Chenu C, manuscript in revision). The use of liraglutide was also investigated in the Goto-Kakizaki T2DM rat model at a dose of 0.4 mg/kg/day for 4 weeks. This regimen led to significant improvement in trabecular and cortical bone microarchitectures in the femur and lumbar vertebra [45].

The effects of liraglutide on bone microarchitecture have also been investigated in a T1DM mouse model. In this study, the administration of 0.093 mg/kg/day liraglutide for 3 weeks did not demonstrate ameliorations of neither trabecular nor cortical microarchitectures [46].

3.3 Effects of GLP-1RAs on tissue material properties

With respect to the improvement in bone strength and intrinsic properties (Young's modulus, stress), that are independent of the bone architecture, one could suspect action of GLP-1RA on tissue material properties. However, very little information has been reported. Tissue material properties represent a set of parameters that describe the modification of biochemical composition or organisation of the bone matrix at the molecular and nanoscale levels [5]. This encompasses for a thorough assessment of the mineral and collagen compartment. Most of our knowledge on the action of GLP-1 on tissue material properties is based on Glp1r KO mice. Indeed, in these animals, a significant reduction in enzymatic collagen cross-linking has been evidenced and associated with alteration of bone strength at the tissue level [37]. However, in opposition to what has been seen with the sister incretin hormone GIP, Glp-1r deletion did not alter the mineral compartment [47]. Data regarding the potential effects of GLP-1RAs on tissue material properties in osteoporotic animals are lacking. However, an elegant study conducted by Mansur et al. investigated the effects of 0.093 mg/kg/day liraglutide over a period of 3 weeks in a T1DM mouse model [46]. These authors reported no amelioration of enzymatic collagen cross-linking or collagen glycation but an unexpected reduction in collagen destruction [46]. Interestingly, we recently showed that collagen maturity was altered in db/db mice and that exenatide treatment at a dose regimen of 10µg/kg/day reversed the impairment in collagen maturity induced by diabetes (Pereira M, Gohin S, Roux JP, Cleasby ME, Mabilleau G, Chenu C, manuscript in revision).

4 GLP-1RAs and blood flow to bone

Diabetes leads to poor circulation and vascular diseases are the principal causes of death and disability in people with diabetes. Consequently, wound and fracture healing are delayed in diabetic patients, one of the main reasons being the impairment in vascularisation [48, 49]. Particularly, diabetes was shown to induce a decrease in endothelial progenitor cells (EPC) that are important for angiogenesis and vascular repair [50]. It is now well established that blood flow is crucial to bone vascular function and osteogenesis [51] and that disrupted blood supply to bone is associated with reduced bone mass, osteonecrosis and impaired bone regeneration [49, 52, 53]. Very little work has however examined whether the bone vasculature and bone blood flow are reduced in diabetic bone and if it is possible to restore them with the use of anti-diabetic drugs. Fajardo [54] recently reviewed the literature

regarding the microvascular complications in diabetic bone but evidences are still lacking to support the link between skeletal fragility in diabetes and those vascular complications.

The potential for GLP-1RAs to enhance vascular function has been demonstrated in a few studies [55-58]. The improvement of vascular endothelial function restores impaired glucose tolerance by ameliorating insulin resistance in skeletal muscle [59]. Interestingly, two weeks administration of 0.5μg/kg/d exenatide was shown to accelerate diabetic wound healing by increasing angiogenesis in the wound and the number of circulating EPCs [60]. Our recent work also demonstrates that 10μg/kg exenatide can have beneficial effects on bone vascularisation in diabetic bone by acutely increasing blood flow to bone in db/db mice (Pereira M, Gohin S, Roux JP, Cleasby ME, Mabilleau G, Chenu C, manuscript in revision). This suggests that the increased bone formation induced by exenatide treatment in diabetic mice could be attributed in part to this increased skeletal perfusion. More work is therefore needed to examine whether skeletal perfusion is linked to bone formation in diabetic bone and if GLP-1RAs could be used as treatment to increase vascularisation in diabetic patients with poor fracture healing.

5 GLP-1 RA and hormones that regulate bone metabolism

A major breakthrough in the bone research field has been the finding that bone is an endocrine organ that can affect other organs via the release of hormones such as osteocalcin and sclerostin. There are increasing reports showing that GLP-1RAs can affect the release of these hormones by bone cells *in vitro* and in animal models but the clinical evidence is however still very scarce.

5.1 Sclerostin

The discovery of the importance of the Wnt/β catenin pathway for bone formation has led to extensive work examining the function of sclerostin in bone. Sclerostin is a product of the *SOST* gene expressed mainly by osteocytes which is secreted and acts as a potent antagonist of Wnt signalling [61]. Its deficiency or its pharmacological neutralisation increases bone formation, making it a potential target for treatment of bone diseases associated with bone loss, such as osteoporosis [62, 63]. Several studies have shown that serum sclerostin levels are elevated in diabetic patients, suggesting that sclerostin could contribute to the decreased bone formation observed in diabetic patients [64-66]. There are however conflicting results regarding the changes in serum sclerostin levels observed

among studies, partly due to the fact that sclerostin could be derived from other non-skeletal sources so that serum levels may not reflect the production in bone [67] and partly because the ELISA kits for sclerostin measurements were found to lack accuracy [68, 69].

To address this issue, a few studies were conducted examining if sclerostin production by osteocytes is modified in bone of diabetic rodents or *in vitro* when osteocytes are cultured in high glucose levels. Inconsistencies were observed [39, 70] and *in vitro* studies are however constrained by the fact that only osteocyte cell lines can be used and they are discrepancies regarding their production of sclerostin [39, 70]. Our recent study shows that the impaired bone microarchitecture and cellular turnover associated with T2DM-like conditions in diabetic ZDF rats are not correlated with changes in serum sclerostin levels, bone sclerostin expression or osteocyte viability [39]. On the other hand, high fat diet in mice resulted in increased serum sclerostin and dramatic alterations of osteocyte network organisation [71]. Few studies have investigated if GLP-1RAs could affect sclerostin production. Although most data agrees that GLP-1r is mainly expressed in immature osteoblasts, GLP-1r can be present in osteocytes where it co-localise with sclerostin [38], suggesting that GLP-1RAs may affect sclerostin production by osteocytes. Kim et al have indeed shown that sclerostin levels are increased in diabetes and can be down-regulated by exenatide treatment [38]. More recently, they demonstrate that the DPP-4 inhibitor vildagliptin lowers the increases levels of sclerostin induced by thiazolidinedione [72]. Our own results are more conflicting as we showed that exenatide but not liraglutide decreased sclerostin levels in OVX mice [31], but in contrast, sclerostin levels were increased with exenatide in diabetic mice while unchanged in IR rats (Pereira M, Gohin S, Roux JP, Cleasby ME, Mabilleau G, Chenu C, manuscript in revision). This may reflect differences in the currently available commercial assays for the measurement of sclerostin or the animal model making presently the measurement of serum sclerostin not very useful as a predictive biomarker for impaired bone formation in T2DM.

5.2 Osteocalcin

Osteocalcin (OC) is a small protein produced in bone by osteoblasts during bone formation which has traditionally been used as a serum marker for bone formation [73]. This protein has however regained a different interest in recent years due to the demonstration that when it is in its uncarboxylated form (GluOC) which does not bind to bone, it can circulate, act as a hormone and regulate glucose metabolism [74]. GluOC can stimulate the release of GLP-1

from the small intestine and therefore indirectly promote insulin secretion by the pancreatic β–cell [75].

It was suggested that incretins could contribute to whole body energy metabolism by modulating osteocalcin synthesis in osteoblasts. The effects of GLP-1RAs on osteocalcin production by osteoblasts were examined and once again the results are inconsistent. While Kim et al [38], Nuche-Berenguer et al [44] demonstrate an increase in serum osteocalcin levels with exendin-4 in T2DM and IR rats, this was not the case with liraglutide treatment [21, 76]. A recent study demonstrates that incretins inhibit thyroid hormone-stimulated osteocalcin synthesis in osteoblasts *in vitro*, suggesting that incretins could stimulate bone formation by reducing the osteocalcin levels [77], although this is not confirmed *in vivo*. Osteocalcin concentration significantly increases during calcification and arterial calcification is an important complication of diabetes due to the differentiation of vascular smooth muscle cells into osteoblast-like cells. Although some work demonstrates an inhibitory effect of GLP-1RAs on vascular calcifications, this is not always the case [78, 79].

5.3 Calcitonin

Calcitonin is a peptide hormone produced by the thyroid parafollicular cells, commonly named "C-cells," that regulate calcium homeostasis [80]. Increases in serum calcium activate the release of calcitonin from the C-cells, which consecutively inhibits bone resorption by the osteoclast and calcium absorption by the intestine. It was therefore one of the first agents to be used as a treatment for osteoporosis. As mentioned previously, several animal studies suggest that GLP-1RAs can affect bone metabolism indirectly via the release of calcitonin by thyroid C cells which express the GLP-1r [26, 81]. The expression of the GLP-1r in thyroid glands has indeed been documented in rodents [82], but there is an uncertainty regarding its expression in humans [83, 84]. Furthermore, basal and stimulated calcitonin did not change during 1 year of liraglutide treatment [85]. Our own work demonstrates that serum levels of calcitonin were indeed increased by exendin-4 treatment in ovariectomised mice [31], although this was not shown when mice were treated with liraglutide, suggesting once again that these two GLP-1 agonists may have a different mechanism of action.

6 Conclusion

Based on several rodent studies, GLP-1 therapy emerges as one of the most promising anti-diabetic therapy for treating the skeletal fragility associated with diabetes. It was shown to increase bone mass, improve trabecular and cortical architectures, enhance bone strength and tissue material properties, affecting the collagen compartment rather than the mineral one. The possible mechanisms of action of GLP-1RAs on the skeleton are illustrated in Figure 1. They are however still not very clear and different GLP-1RAs may have different means of action. Among the potential ones, the stimulation of bone blood flow by GLP-1RAs seems very interesting and extremely promising in situations of osteoporotic and diabetic fractures. Clinical data are however still lacking and those establishing the relationship between the GLP-1RA use and decrease fracture risk have been so far negative. There is therefore a need for long-term clinical studies comparing the skeletal effects of different GLP-1RAs.

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Table 1: Summary of approved GLP-1RAs for the treatment of type 2 diabetes mellitus

Active compound	Drug name	Marketed by	Approved in	Approved dose range
Exenatide (or Exendin-4)	Byetta	Astra Zeneca AB	2006	5-10 µg twice daily
Liraglutide	Victoza	Novo Nordisk A/S	2009	0.6-1.8 mg once daily
Lixisenatide	Lyxumia	Sanofi Aventis Groupe	2013	10-20 µg once daily
Exenatide (or Exendin-4) long acting release	Bydureon	Astra Zeneca AB	2011	2 mg once weekly
Albiglutide	Eperzan	GlaxoSmithKline Trading Services Ltd	2014	30-50 mg once weekly
Dulaglutide	Trulicity	Eli Lilly Nederland B.V.	2014	0.75-1.5 mg once weekly

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Figure 1: Simplified scheme of the possible skeletal effects of GLP-1RAs