FGFR antagonists restore defective mandibular bone repair in a mouse model of osteochondrodysplasia

Anne Morice[1], Amélie de la Seiglière[1], Alexia Kany[1], Roman H Khonsari[1], Morad Bensidhoum[2], Maria-Emilia Puig-Lombardi[3], Laurence Legeai Mallet[1]

1. Université de Paris Cité, Imagine Institute, Laboratory of Molecular and Physiopathological Bases of Osteochondrodysplasia, INSERM UMR 1163, Paris
2. B3OA UMR CNRS 7052, Université Paris Cité, Paris
3. Bioinformatics Core Platform, Imagine Institute, INSERM UMR1163 and Structure Fédérative de Recherche Necker, INSERM US24/CNRS UAR3633, Université Paris Cité, Paris

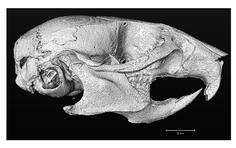
Background

Hypochondroplasia (HCH) is linked to Fibroblast Growth Factor Receptor 3 (*FGFR3)* gain-of-function mutations. These patients are characterized by **rhizomelic dwarfism**, and **craniomaxillofacial anomalies** requiring craniomaxillofacial bone surgeries. Our team generated the **first *Hch* mouse model (*Fgfr3*$^{N534K/+}$)** (Loisay et al, 2023), expressing the most common missense FGFR3 mutation (p.Asn540Lys), presenting progressive dwarfism, maxillary retrusion and mandibular prognathism mimicking human pathology.

Previously we demonstrated that **mandibular bone healing was impaired in this *Hch* mouse model**, we observed a **defective bone consolidation, significant microarchitectural alterations of the calluses** and abnormal presence of **pseudarthrosis** (Morice *et al*, Bone Research, 2025).

Objectives

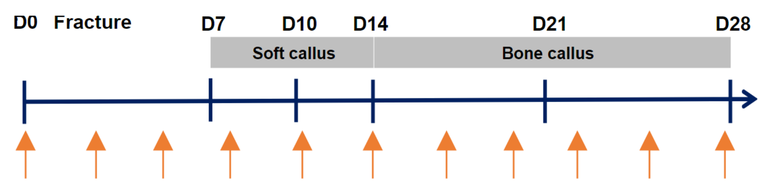
Analyzing **the effects of two FGFR3 antagonists on mandibular bone repair** in this *Hch* mouse model:

***Fgfr3*$^{+/+}$**   ***Fgfr3*$^{N534K/+}$ (HCH model)**

1) **a tyrosine kinase inhibitor, Infigratinib (BGJ398)** (phase II clinical trial for achondroplasia children) ;
2) **vosoritide, a CNP analog (BMN 111)** (Voxzogo®) with FDA and EC approval for achondroplasia children.

Methods

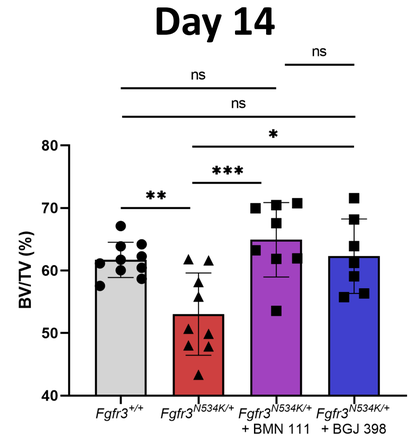
We performed **unstabilized fractures** at 6 weeks of age on the right mandible in *Hch* mice.

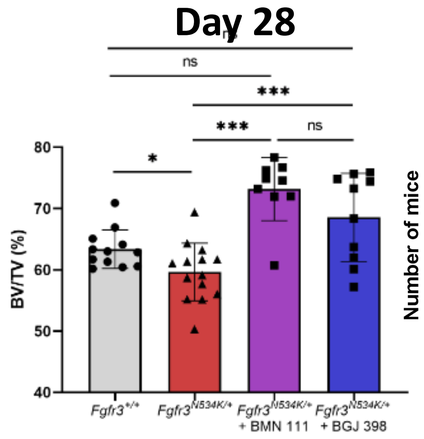


At day 0 post fracture, **subcutaneous injections** were performed with **BGJ398 (4 mg/kg)** or **BMN111 (0.8 mg/kg)** 3 times/week, during either 14 or 28 days. **Morphometric analyses** concerned two major key points of bone repair, D14 and D28 post fracture.

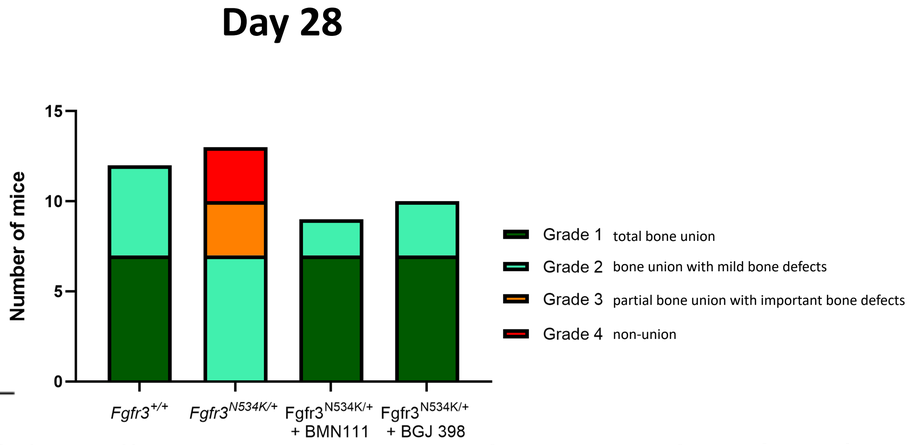
Results

BV/TV

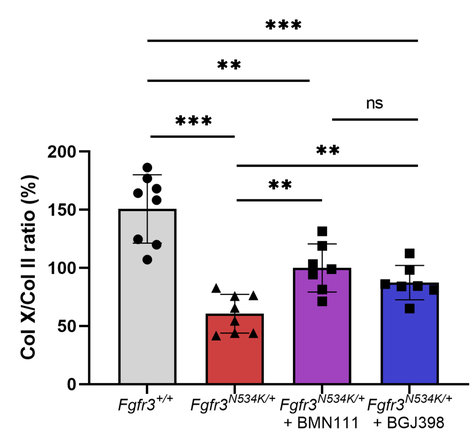




Consolidation grades



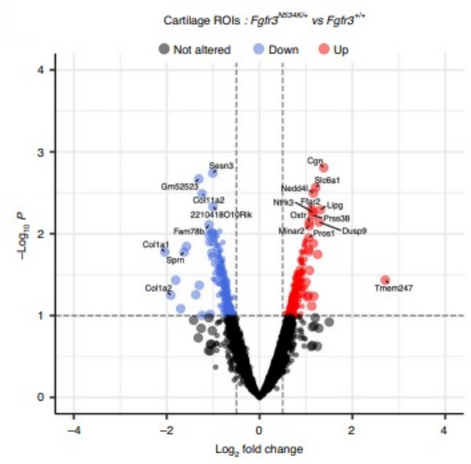
Chondrocyte differentiation (callus D14)

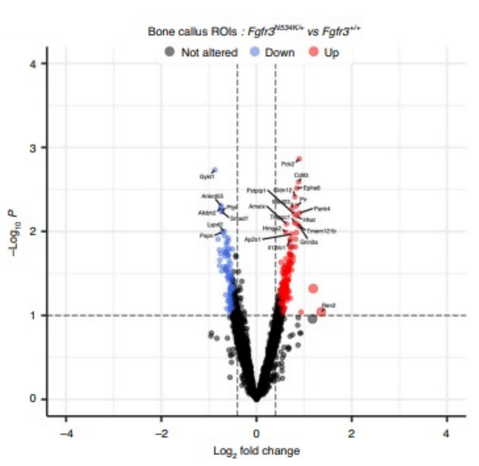


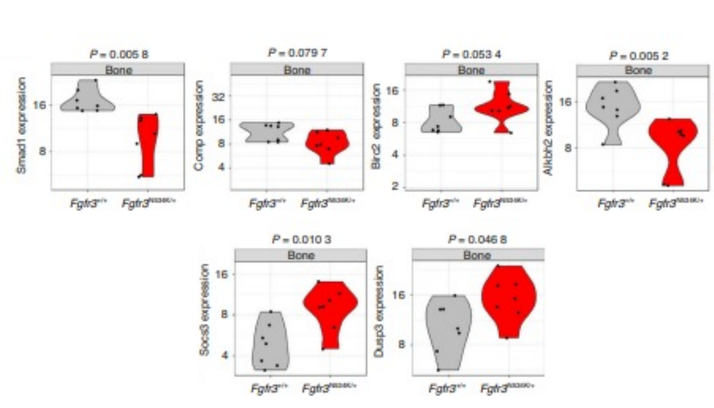
Bone Volume/Total Volume (**BV/TV**) values of the calluses **were improved in treated groups: D14 (+29 %, p<0.005** (BMN 111), **+20.8 %, p<0.05** (BGJ 398)); **D28 (+ 24%, p<0.005** BMN 111), (**+ 17,8%, p<0.01** BGJ 398). **Calluses volumes** were significantly reduced at D28: **-30.9 %, p<0.005** (BMN 111); **- 38,6 %, p<0.0001** (BGJ 398), corresponding to **accelerated bone remodeling**. **All treated mutants presented normal grades of consolidation (grades 1 or 2), without pseudarthrosis** (grade 4) at D28.

Chondrocyte differentiation was significantly improved by both treatments: significant increase of the ratio of **Col X / ColII** chondrocyte positive area : BMN111: **+64.9 %, p<0.01**; BGJ398: **+40.6 %, p<0.01** in *Fgfr3*$^{N534K/+}$ mice compared to control mice.

Spatial transcriptomic analysis of the cartilage and newly formed bone in the calluses from *Fgfr3*$^{N534K/+}$ and *Fgfr3*$^{+/+}$ mice at day 14 post-fracture







Volcano plot representations of differentially expressed genes (*P*<0.1) that were either **upregulated (in red) or downregulated (in blue) in the cartilage of the calluses** in *Fgfr3*$^{N534K/+}$ mice, relative to *Fgfr3*$^{+/+}$ mice.

Violin plots showing the **expression levels** (normalized read counts) of ***Col11, Col1a1, Dmp1, Phospho1, Hmgb2, Kif17*** and ***Dusp9*** **in the cartilage of the calluses** from *Fgfr3*$^{N534K/+}$ mice (in red) and *Fgfr3*$^{+/+}$ mice (in grey).

Volcano plot representations of differentially expressed genes (*P*<0.1) that were either **upregulated (in red) or downregulated (in blue) in the bone of the calluses** in *Fgfr3*$^{N534K/+}$ mice, relative to *Fgfr3*$^{+/+}$ mice.

Violin plots showing the **expression levels** (normalized read counts) of ***Smad1, Comp, Birc2, Alkbh2, Dusp3*** and ***Socs3*** in the newly formed bone of the calluses from *Fgfr3*$^{N534K/+}$ mice (in red) and *Fgfr3*$^{+/+}$ mice (in grey).

Conclusions and perspectives

Our findings suggest that **both MAPK and STAT pathways are over-activated in the bone callus in a mouse model of hypochondroplasia,** and thus highlight the key role of these downstream signaling pathway in FGFR3-related disorders.

Our data showed that the **constitutive activation of FGFR3 was counteracted by infigratinib and vosorotide thus restoring the defective mandibular endochondral bone repair**, this modulation of FGFR3 signaling might be of value for treating bone repair in patients with FGFR3 chondrodysplasia.