Integrated analysis of long-term growth and bone development in pediatric and adolescent patients receiving bevacizumab

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Abbreviations

BMI	body mass index
EFS	event-free survival
NCI	National Cancer Institute
SDS	standard deviation score
VEGF	vascular endothelial growth factor

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Abstract

Background: We conducted an integrated analysis of clinical data to describe long-term effects of bevacizumab on growth and bone development in pediatric and adolescent patients with solid tumors.

Procedure: Clinical data were pooled from five phase I/II trials of bevacizumab versus chemotherapy: BERNIE, HERBY, and AVF4117s enrolled newly diagnosed patients, AVF3842s and AVF2771s enrolled patients with relapsed/refractory disease. Height, weight, body mass index (BMI), and bone-age data were pooled by treatment group. Growth charts were used to track and monitor growth in relation to a reference population of healthy children. Bone age was measured based on X-ray of the left hand and wrist. Analyses were exploratory/descriptive.

Results: Overall, 268 patients received bevacizumab ± chemotherapy and 135 received chemotherapy alone. Baseline characteristics were generally balanced. Median duration of long-term follow-up was 41.8 months (range, 2.4–75.1) with bevacizumab and 22.9 months (range, 2.8–69.2) with chemotherapy alone. Patients had age-appropriate baseline height and weight. Mean height and weight percentiles decreased over time in both treatment groups, but remained within the normal range (height: mean standard deviation score [SDS] range -2 to +3; weight: mean SDS range -2 to +1). Similar trends were seen in BMI. A tendency for reduced growth velocity relative to the reference population was observed at 6 months and 1 year in both groups, but there was no additional decrease for patients receiving bevacizumab.

Conclusion: Bevacizumab did not appear to have additional negative effects on growth or development of pediatric and adolescent patients with solid tumors.

1 | INTRODUCTION

The anti-vascular endothelial growth factor (VEGF) antibody, bevacizumab, is approved for use in combination with chemotherapy in a number of adult tumors and has a well-established safety profile in adults.[1,2] However, there are limited data on the effects of bevacizumab in pediatric and adolescent patients with cancer.

A correlation between high serum VEGF levels and adverse prognostic outcome has been reported in several preclinical and pilot studies in pediatric neuroblastoma, osteosarcoma, Ewing sarcoma, Wilms tumor, and rhabdomyosarcoma,[3–9] suggesting that anti-VEGF agents may be a useful therapeutic approach in these patients. However, in two phase II trials, the addition of bevacizumab to standard therapy for children and adolescents with untreated metastatic soft tissue sarcoma[10] or osteosarcoma[11] did not significantly improve event-free survival (EFS); its safety profile was consistent with the known safety profile in adults. Similarly, in a phase II trial in pediatric patients with newly diagnosed high-grade glioma, the addition of bevacizumab to radiotherapy-temozolomide failed to prolong EFS.[12] Sustained disease control was, however, reported with bevacizumab and irinotecan in children with recurrent low-grade gliomas in an earlier phase II study.[13]

Angiogenesis and VEGF play a key role in bone growth and development.[14,15] Abnormalities in growth plates have been identified in animals treated with anti-VEGF agents, including bevacizumab.[16,17] In juvenile cynomolgus monkeys with open growth plates, severe physeal dysplasia was observed following bevacizumab treatment at up to 20 times the recommended human dose.[18] Pregnant rabbits dosed with up to 12 times the human bevacizumab dose during gestation displayed reduced or irregular ossification in the skull, jaw, spine, ribs, and tibia, as well as

decreased maternal and fetal body weight.[18] In addition, in a combined analysis of children with refractory cancer receiving antiangiogenic therapies in six phase I trials, a small but relevant proportion of patients (9.4%) experienced growth plate abnormalities, though none were receiving bevacizumab.[19]

Since little is known about the long-term effects of bevacizumab on growth and bone development in pediatric cancer patients, we conducted an integrated analysis of clinical trial data to describe these effects in children and adolescents with solid tumors.

2 | METHODS

2.1 | Study design

As part of a post-marketing commitment, data were pooled for patients aged <18 years who received at least one dose of bevacizumab in one of five phase I/II clinical studies: two randomized, controlled, Roche-sponsored international studies (BERNIE [NCT00643565][10] and HERBY [NCT01390948][12]), two National Cancer Institute (NCI)-sponsored studies (AVF3842s [NCT00381797][13] and AVF2771s [NCT00085111][20]), and a single-arm, investigator-sponsored trial (AVF4117s [NCT00667342][11,21]; Table 1). BERNIE, HERBY, and AVF4117s were conducted in newly diagnosed patients with solid tumors, while the NCI-sponsored studies enrolled patients with relapsed or refractory disease. Study designs and methodology have been published.[10–13,20,21]

Although tumor type, duration of treatment, and study design differed, the parameters assessed in each of the studies were deemed relevant for this analysis. For parameters that were assessed in more than one study, data were pooled to

increase patient numbers and provide a more meaningful analysis; data were not pooled for bevacizumab exposure or parental height as data points were scarce.

The study protocols were approved by applicable ethics committees and institutional review boards, and the studies were conducted in accordance with the Declaration of Helsinki and Good Clinical Practice. Written informed consent was obtained from parents, patient, or legally acceptable representative prior to any study-related procedures.

2.2 | Growth and development

Height and body weight data were collected in all five studies. Epiphyseal maturation and bone age were assessed by a radiologist based on X-ray of the left hand and wrist using the Greulich-Pyle method[22]; these measurements were required in BERNIE, HERBY, and AVF2771s.

Growth charts were used to track a patient's growth over time and to monitor their growth in relation to a reference population of healthy children. World Health Organization growth standards[23] (patients <2 years) and Centers for Disease Control growth reference values[24] (patients ≥2 to 20 years) were used as reference values. A patient's percentile on the growth chart indicated the percentage of the reference population that their value equalled or did not reach for a given growth parameter. A patient's standard deviation score (SDS) indicated to what extent their value deviated from the median of the reference population.

The integrated, descriptive analyses of growth and development included a number of parameters. Height, weight, and body mass index (BMI) were measured versus chronological age. Bone age versus chronological age was assessed before, during, and after treatment, including follow-up. Growth velocity in cm/year was

derived as: (height at time t_2–height at time t_1)/(time between measurements), where time between measurements had to be ≥6 months. All growth and development parameters were assessed separately in males and females from baseline to 6, 12, 24, and 36 months.

Due to the small number of patients with available parental height measurements, prediction methods that require these measurements could not be used for the pooled analysis of genetic growth potential. The Bayley-Pinneau method[25] was therefore used, which is based on bone-age assessment derived from the Greulich-Pyle method[22] that compares X-rays of the left hand and wrist with atlas standards. The assessed sex-specific bone age and its deviation from chronological age is used to predict adult final height by the Bayley-Pinneau method. Bone-age data collected in this way were pooled from BERNIE, HERBY, and AVF2711s; the predicted final height at baseline was compared with the predicted final height during treatment. For those patients with parental height measurements, mid-parental height (the mean of the patient's parents' heights, plus a correction factor for the patient's sex: +6.5cm for males, -6.5cm for females) was compared with the predicted adult final height according to the Bayley-Pinneau height prediction method.[25]

2.3 | Subgroup analysis

The effect of bevacizumab on growth velocity in the growth hormone-dependent phase between infant/toddler and pubescent growth periods, when growth velocity is assumed to be relatively linear, was assessed. Patients aged ≥2 years with a Tanner stage <2 for breast/genitalia and pubic hair were included. If Tanner stage was not available for breast/genitalia or pubic hair, and menarchal status (for females) was

179 missing, females aged ≥2 to <8 years and males aged ≥2 to <9 years were
180 considered to be Tanner stage 1 and therefore pre-pubertal.[26]
181

182 2.4 | Statistical methodology

183 Analyses were exploratory and descriptive and did not have statistically sufficient
184 power. For pooled analyses, all patients who received at least one dose of
185 bevacizumab were assigned to the experimental arm (bevacizumab ± chemotherapy)
186 and all other patients were assigned to the control arm (chemotherapy alone). The
187 pooled control arm included only patients from HERBY and BERNIE, as the
188 remaining studies did not include a control arm.

189 Due to the different schedules in the individual studies, not all assessments
190 were performed at the same timepoint. To pool the assessments for analyses over
191 time, the timepoints were standardized. For all growth assessments except bone
192 age, the timepoints were standardized to 6-month intervals. Any assessment not
193 taken at this timepoint was assigned to the nearest standardized timepoint
194 ± 3 months. Baseline was considered the nearest assessment prior to (up to 2
195 months before or 1 month after) the first study treatment administration.

196 For bone-age assessments and predicted adult height, timepoints were
197 standardized to baseline, during treatment, end of treatment, end of treatment plus 1
198 year, and end of treatment plus 2 years. Baseline was considered the nearest
199 assessment to (up to 14 days before or 30 days after) the first study treatment
200 administration. End-of-treatment timepoint varied by patient. If bone age was not
201 assessed at the end-of-treatment visit, the nearest assessment within 60 days was
202 used. End of treatment plus 1 year (or 2 years) assessment was taken within ± 3
203 months of 1 year (or 2 years) after the end-of-treatment visit.

3 | RESULTS

3.1 | Patients

Overall, 268 patients received bevacizumab ± chemotherapy and 135 patients received chemotherapy alone. Baseline characteristics were generally balanced between the two groups, but the bevacizumab group had fewer adolescents than the chemotherapy group and therefore the median height was significantly lower in the bevacizumab group (although correction for multiple testing would make this difference non-significant; Table 2). Median baseline age was 10.1 years (range, 1–18) in the bevacizumab ± chemotherapy group and 11.0 years (range, 1–17) in the chemotherapy-alone group. The largest proportion of patients was in the growth hormone-dependent phase between infant/toddler and pubescent growth periods (36.9% [n = 99] bevacizumab vs. 37.0% [n = 50] chemotherapy).

3.2 | Previous and concomitant conditions

Considerably more patients in the bevacizumab ± chemotherapy group (60.1% [n = 161]) than in the chemotherapy-alone group (22.2% [n = 30]) had a previous or concomitant disease known to affect growth and fertility (Supplementary Table S1). This was driven by a higher proportion of patients with events in the system organ class ‘neoplasms benign, malignant and unspecified’ (51.5% [n = 138] bevacizumab vs. 2.2% [n = 3] chemotherapy) and in the preferred term ‘neurofibromatosis’ (22.4% [n = 60] bevacizumab vs. 1.5% [n = 2] chemotherapy), which can lead to delayed or early puberty, and small stature.[27] The higher incidence of recorded ‘previous or concurrent neoplasms’ in the bevacizumab ± chemotherapy group is most likely explained by the fact that a large proportion of these patients (37.3% [n = 100]) came

from studies AVF2771s and AVF3842s, which enrolled patients with refractory or recurrent disease, whereas all patients in the chemotherapy-alone group came from BERNIE and HERBY, which included patients with newly diagnosed disease. The higher incidence of neurofibromatosis in the bevacizumab ± chemotherapy group is due to the fact that 58/60 patients enrolled in study AVF3842s had neurofibromatosis. This study only included a bevacizumab arm and was restricted to patients with refractory or recurrent low-grade glioma. There was also a higher incidence of craniospinal irradiation in the bevacizumab ± chemotherapy group (19.8% [n = 53]) versus the chemotherapy-alone group (0%), which was driven by the high frequency of previous/concomitant radiation in patients with recurrent or refractory gliomas in study AVF3842s.

Most patients had received at least one previous or concomitant medication known to affect growth, other than corticosteroids (most frequently chemotherapy): 67.2% (n = 180) in the bevacizumab group and 62.2% (n = 84) in the chemotherapy group (Supplementary Table S2). The majority of patients had also received at least one previous or concomitant corticosteroid, but the proportion was lower among patients receiving bevacizumab (68.7% [n = 184]) versus chemotherapy alone (82.2% [n = 111]). Most patients who received corticosteroids took them for more than 7 consecutive days (41.4% [n = 111] bevacizumab vs. 54.8% [n = 74] chemotherapy).

3.3 | Treatment exposure

The number of bevacizumab administrations per patient, as well as the average dose per administration, differed across the five studies in line with exposures planned in the individual protocols. The mean number of bevacizumab

administrations per patient ranged from 5.6 to 19.9 in the individual studies, and the dose of bevacizumab across the studies ranged from 5 to 15 mg/kg every 2 or 3 weeks (Table 1). The median duration of long-term follow-up from enrollment across the studies was 3.5 years (range, 0.2–6.3) in the bevacizumab ± chemotherapy group and 1.9 years (range, 0.2–5.8) in the chemotherapy-alone group.

3.4 | Height, weight, and BMI

At baseline, children in both treatment groups had age-appropriate height and weight, which was similar to the reference population (i.e., the mean SDS was close to 0): mean SDS for height: -0.01 bevacizumab, +0.15 chemotherapy; mean SDS for weight: +0.33 bevacizumab, +0.18 chemotherapy. Mean height and weight percentiles generally decreased over time, more so for the chemotherapy-alone group (Fig. 1) and, although lower than the reference population, remained within the normal range at all timepoints: mean SDS for height ranging from -2 to +3; mean SDS for weight ranging from -2 to +1.[22] Similar trends were seen in BMI over time.

3.5 | Bone age

Bone-age assessments were pooled from studies with available data (BERNIE, HERBY, and AVF2771s) at baseline (n = 231), end of treatment (n = 68), end of treatment plus 1 year (n = 27), and end of treatment plus 2 years (n = 10), as the treatment duration of the individual studies differed. There was no indication of any difference in bone age compared with chronological age in patients receiving bevacizumab ± chemotherapy versus chemotherapy alone, regardless of age (Fig. 2). In both treatment groups, bone age for the majority of patients was within the normal range (± 1 year) at all timepoints.

3.6 | Growth velocity

A tendency for reduced growth velocity relative to the reference population was observed at 6 months and 1 year in both treatment groups (when most patients were receiving study treatment) in females (Fig. 3) and males (Fig. 4). No clear growth spurt was observed in the pre-pubertal period. Regardless of age, sex, and timepoint, there was no indication of an additional decrease in growth velocity for patients receiving bevacizumab alone or in combination with chemotherapy compared with those receiving chemotherapy alone. Caution should be used in the interpretation of the results due to the limited patient numbers at later timepoints: 6 months (n = 166 males, n = 136 females), 1 year (n = 113 males, n = 92 females), 2 years (n = 39 males, n = 38 females), 3 years (n = 25 males, n = 22 females).

3.7 | Subgroup analysis

The analyses of growth and development were performed in the HERBY and BERNIE randomized studies separately, and the results were consistent with the pooled analysis (data not shown).

Analyses of height (Supplementary Fig. S1), bone age (Supplementary Fig. S2), and growth velocity (Supplementary Fig. S3 and S4) were conducted in patients in the growth hormone-dependent phase (females ≥2 to <8 years, males ≥2 to <9 years). Results were consistent with the overall patient population, with no indications of additional negative effects for patients receiving bevacizumab ± chemotherapy versus chemotherapy alone.

3.8 | Genetic growth potential

Genetic growth potential was assessed using data from HERBY, in which the collection of parental height was included by a protocol amendment. Genetic growth potentials were similar between patients with available data receiving bevacizumab ± chemotherapy (n = 7) and chemotherapy alone (n = 4). Patients' genetic growth potentials were compared with their predicted adult heights according to Bayley-Pinneau.[22,25] Due to the limited data, no conclusions could be made regarding the difference between patients' genetic growth potentials and their predicted adult heights over the course of the study. However, in both treatment arms, patients' predicted median adult heights at baseline were already higher than their genetic growth potentials, which could indicate that the method of prediction overestimated actual genetic growth potentials.

4 | DISCUSSION

Our analysis investigated the effects of bevacizumab on long-term growth and bone development in pediatric and adolescent patients with cancer, with median duration of follow-up in patients receiving bevacizumab of 3.5 years. Although lower than the reference population, height, weight, BMI, and bone age for both treatment groups remained within the normal range at all timepoints. Given the poor clinical status of the patients, it was to be expected that their height, weight, and BMI would be lower than the reference population. Growth velocity was also lower than in the reference population in both sexes. Importantly, however, there was no differentiation between patients who received bevacizumab ± chemotherapy or those who received chemotherapy alone. These results were consistent for patients in the growth hormone-dependent phase and in patients with longer-term follow-up. The small

difference in median baseline height between the two patient groups was not expected to have influenced the results.

Growth, bone development, and epiphyseal maturation are of concern in pediatric patients receiving antiangiogenic agents such as bevacizumab.[19] However, few publications have reported safety data on the use of bevacizumab in children and adolescents.[13,19,20,28–31] Growth plate abnormalities were evaluated in a combined analysis of six phase I trials in children with different tumors evaluating new antiangiogenic therapies. While most patients had no evidence of growth plate toxicity, five patients (9.4%) had epiphyseal abnormalities.[19] One of the five patients also experienced progressive epiphyseal widening. However, this patient met height expectations following cessation of the antiangiogenic therapy, and the cartilage magnetic resonance imaging sequences resolved, suggesting that epiphyseal changes may be reversible.

We experienced a number of difficulties with data collection and method standardization. Protocol amendments were put in place during BERNIE, HERBY, and AVF2771s to ensure sufficient growth and development measurements would be collected, and investigators were prompted to complete protocol-mandated assessments. Despite these efforts, a number of growth and height measurements were incomplete. The intensity of the treatment, and the poor performance status or early progression of the patients, meant that these measurements were not consistently collected at baseline or follow-up. Although some data points were available up to 66 months, beyond 36 months the patient numbers became very small, therefore limiting the conclusions that could be drawn.

Ideally, genetic potential should be compared with actual results to check for diminished growth in pediatric patients. However, parental height data were not

collected in four of the studies and it was not deemed feasible to do this analysis retrospectively. Following a protocol amendment, these data were limited to patients randomized late in HERBY and to those surviving and consenting at the time of the amendment. Furthermore, bone-age measurements were not consistently collected at baseline or at follow-up in BERNIE and HERBY, meaning that critical data points were missing. Where feasible, retrospective bone-age X-rays were collected for surviving patients, but there was no central review of these data. Given the limited control regarding data collection in studies AVF2771s, AVF3842s, and AVF4117s, it is unknown whether a stadiometer was used consistently for height measurement in all patients. It is assumed that individual patient height was consistently assessed during each study, which enabled an assessment of deviations over time.

This integrated analysis has some limitations, in that not all of the studies were randomized, and the different protocols were not designed to collect data for the same growth and development endpoints. As this was a retrospective evaluation of the databases of five individual studies, data cleaning was not possible for the data set as a whole.

In summary, the large cohort of more than 400 pediatric and adolescent patients enabled a good assessment of the long-term effects of bevacizumab, although sample sizes were smaller at later timepoints as many of the patients did not survive to contribute follow-up data.

5 | CONCLUSION

Acknowledging the limitations of this analysis, we found no apparent negative effects of bevacizumab on growth and development in pediatric and adolescent patients

who received bevacizumab ± chemotherapy compared with those who received chemotherapy alone.

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CONFLICT OF INTEREST STATEMENT

H.L.M. has received travel, accommodation, or expenses from Ipsen Pharma. J.G. has received honoraria, research funding, and travel, accommodation, or expenses from Roche, Novartis, and Bristol-Myers Squibb; and has acted in a consulting or advisory role to Roche, Novartis, and Bristol-Myers Squibb. D.H. has received honoraria from AstraZeneca, Bayer, Boehringher Ingelheim, GlaxoSmithKline, Merck, Novartis, and Roche; acted in a consulting or advisory role to AbbVie, AstraZeneca, Bayer, Boehringher Ingelheim, Bristol-Myers Squibb, Celgene, GlaxoSmithKline, Merck, Novartis, Pfizer, and Roche; received research funding from AstraZeneca; and received travel, accommodation, or expenses from AstraZeneca, Bayer, Boehringher Ingelheim, Bristol-Myers Squibb, GlaxoSmithKline, Novartis, and Roche. J.G.B. has received research funding from Amgen, Bristol-Myers Squibb, Celgene, Eisai, Ignyta, Lilly, Merck, Novartis, and Pfizer. S.G. has received honoraria from Celgene Corporation and BioMarin Pharmaceuticals; acted in a consulting or advisory role to Celgene Corporation and BioMarin Pharmaceuticals; and received research funding and travel, accommodation, or expenses from Celgene Corporation and Bristol-Myers Squibb. M.J. is employed by Genentech Inc.; owns stock in Roche; and has received travel, accommodation, or expenses from Genentech Inc. J.B. is employed by F. Hoffman La-Roche Ltd.

M.C.E. is employed by F. Hoffman La-Roche Ltd and owns stock in Roche. S.F.-R. is employed by F. Hoffman La-Roche Ltd.; owns stock in Roche; and has received travel, accommodation, or expenses from Roche. J.H.M.M., B.G., and F.N. have no conflicts of interest to disclose.

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FIGURE LEGENDS

FIGURE 1 Standard deviation score (SDS) for patient height over time for (A) bevacizumab ± chemotherapy and (B) chemotherapy alone, and patient weight over time for (C) bevacizumab ± chemotherapy and (D) chemotherapy alone. Dashed lines represent the median height and weight of the reference population. Approximately 95% of the reference population would be expected to have a SDS between -2 and 2. Blue dots indicate the median for the patient population while the whiskers indicate the range of the data. BL, baseline

FIGURE 2 Bone age versus chronological age at (A) baseline, and (B) end of treatment plus 1 year. The identity line shows where bone age equals chronological age

FIGURE 3 Scatter plot of growth velocity in female patients at various times post-baseline: (A) 6 months, (B) 1 year, (C) 2 years, and (D) 3 years. Solid curves represent growth velocity of the reference population. Non-linearity of the curves is for technical reasons or due to the change in reference standard

FIGURE 4 Scatter plot of growth velocity in male patients at various times post-baseline: (A) 6 months, (B) 1 year, (C) 2 years, and (D) 3 years. Solid curves represent growth velocity of the reference population. Non-linearity of the curves is for technical reasons or due to the change in reference standard