A mammalian model for Laron syndrome produced by targeted disruption of the mouse growth hormone receptor/binding protein gene (the Laron mouse)

Yihua Zhou*[†], Bixiong C. Xu*, Hiralal G. Maheshwari[‡], Li He*[†], Michael Reed*, Maria Lozykowski*, Shigeru Okada*, Lori Cataldo*, Karen Coschigamo*, Thomas E. Wagner*, Gerhard Baumann[‡], and John J. Kopchick*[§]

*Edison Biotechnology Institute, Molecular and Cellular Biology Program, and Department of Biomedical Sciences, College of Osteopathic Medicine, Ohio University, Athens, OH 45701; and ‡Center for Endocrinology, Metabolism and Molecular Medicine, Department of Medicine, Northwestern University Medical School, Chicago, IL 60611

ABSTRACT Laron syndrome [growth hormone (GH) insensitivity syndrome] is a hereditary dwarfism resulting from defects in the GH receptor (GHR) gene. GHR deficiency has not been reported in mammals other than humans. Many aspects of GHR dysfunction remain unknown because of ethical and practical limitations in studying humans. To create a mammalian model for this disease, we generated mice bearing a disrupted GHR/binding protein (GHR/BP) gene through a homologous gene targeting approach. Homozygous GHR/BP knockout mice showed severe postnatal growth retardation, proportionate dwarfism, absence of the GHR and GH binding protein, greatly decreased serum insulin-like growth factor I and elevated serum GH concentrations. These characteristics represent the phenotype typical of individuals with Laron syndrome. Animals heterozygous for the GHR/BP defect show only minimal growth impairment but have an intermediate biochemical phenotype, with decreased GHR and GH binding protein expression and slightly diminished insulin-like growth factor I levels. These findings indicate that the GHR/BP-deficient mouse (Laron mouse) is a suitable model for human Laron syndrome that will prove useful for the elucidation of many aspects of GHR/BP function that cannot be obtained in humans.

Laron syndrome, a recessively inherited disease with growth hormone (GH) resistance, was first described in 1966 (1). To date, more than 220 cases have been reported worldwide. Affected patients are characterized by very short stature, facial dysmorphism, truncal obesity, delayed puberty, recurrent hypoglycemia, low serum insulin-like growth factor I (IGF-I), elevated serum GH, absent, low, or dysfunctional serum GH binding protein (GHBP), and resistance to GH (2). The primary defect causing this disease is a mutated GH receptor (GHR). Absence of GH binding to GHRs in the liver has been demonstrated in two patients (3). A spectrum of ≈30 inactivating mutations affecting the expression or functions of the GHR and GHBP has been reported (2, 4–8). They include deletions, nonsense, frameshift, missense, and splice site mutations, mostly within the extracellular domain of the GHR, resulting in truncations, abnormal processing, or inability to bind ligand. Other mutations disable GHR dimerization or truncate the intracellular portion, with resulting inability to transduce a signal (9, 10). The phenotype of Laron syndrome is primarily defined on clinical and blood biochemical grounds because of ethical and practical limitations in defining tissue GHR status and its biology in humans. Several other issues

related to GH resistance, such as long term IGF-I responsivity, longevity, immune function, oncogenesis, etc., are only partially known because of the slow growth phase and long life-span of humans. A suitable animal model would greatly facilitate a full investigation of the consequences of GHR dysfunction. However, to date, only one animal with a mutated GHR, the sex-linked dwarf chicken (11), has been discovered, and no mammalian model exists for the human disorder. The dwarf chicken is not an ideal paradigm for a human disease because of the significant taxonomic differences between birds and mammals. To create a mammalian model for Laron syndrome, we disrupted the mouse GHR/BP gene by homologous recombination and studied the growth and endocrine status of the resultant GHR/BP-deficient mice (Laron mice).

The GHR and GHBP are encoded by a single GHR/BP gene in mammalian species (4, 12, 13). The GHR is a single chain glycoprotein whereas the GHBP is a secreted short form that corresponds to the extracellular domain of the GHR (12). GHBP is generated either by proteolysis from the GHR (humans, rabbits, and pigs) (12, 14, 15) or through alternative splicing of a common GHR/BP pre-mRNA (rodents) (13, 16–18). The GHR and GHBP are expressed in virtually all tissues (19–21). The existence of other types of GHRs has been postulated (22) but thus far none has been identified.

MATERIALS AND METHODS

Gene Disruption Strategy. We aimed to disrupt the mouse (m)GHR/BP gene by homologous recombination. The GHR/BP is encoded in both humans and mouse by 10 exons (4, 13), among which exon 4 encodes a portion of the GH binding domain (23). To produce the GHR/BP null mutation, a 500-bp *Dra*III fragment containing the 3′ portion of exon 4 and the adjacent 5′ region of intron 4/5 was replaced by a *neo* cassette (Fig. 1*A*). The rationale for disrupting the gene at exon 4 is 2-fold: (*i*) mutations in exon 4 have been reported in patients with Laron syndrome (4–7), and (*ii*) if alternative pre-mRNA splicing should unexpectedly occur between exons 3 and 5, the resulting frameshift would result in a premature stop codon in exon 5 (4, 13).

Construction of a GHR/BP Targeting Vector. A 15-kb *Eco*RI fragment containing exon 4 of the mGHR/BP gene was isolated
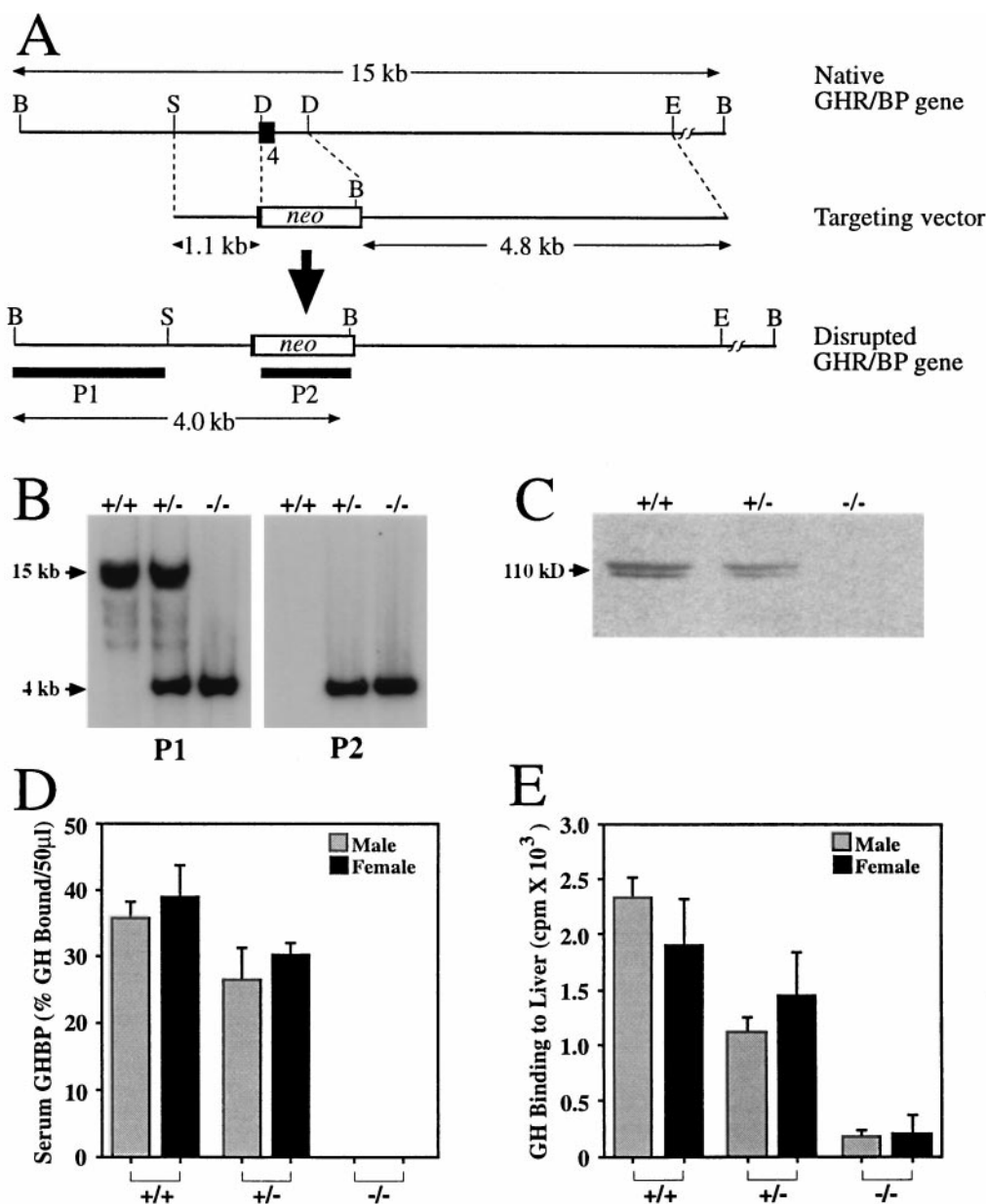

FIG. 1.   Targeted disruption of the mGHR/BP gene. (*A*) Strategy for gene disruption. To construct the targeting vector, a *Dra*III–*Dra*III fragment containing a major portion of GHR exon 4 and ≈500 bp of intron 4/5 was replaced by the neomycin resistance (*neo*) gene. A herpes simplex virus thymidine kinase gene cassette (not shown) was placed at the 3′ end of the targeting vector for negative selection. The lengths (kilobases) of the left and right arms of the targeting vector and the important restriction fragments are indicated in the figure. B, *Bam*HI; D, *Dra*III; E, *Eco*RI; and S, *Sac*I. P1 and P2 denote hybridization probes. (*B*) Mouse genotyping with Southern blots using genomic DNA isolated from GHR/BP$^{+/+}$, GHR/BP$^{+/-}$, and GHR/BP$^{-/-}$ mice, digested with *Bam*HI, and hybridized with ^{32}P-labeled probes P1 or P2. The size of the native *Bam*HI band was 15 kb, and that of the recombinant fragment was 4 kb. (*C*) Western blot analysis of liver GHR in a 4–12% gradient SDS/PAGE gel. The doublet appearance of the GHR is likely caused by variations in glycosylation. (*D*) GHBP activity in sera collected at 30 and 60 days of age (*n* = 6–7 mice in each group; data from 30- and 60-day-old animals pooled because they were not significantly different). No GHBP was detected in GHR/BP$^{-/-}$ mice (<30 pM). (*E*) Specific binding of [^{125}I]-bGH to liver membrane preparations from 60-day-old mice (*n* = 9 animals in each group).

from a C57/Black mouse genomic library (13). To construct a GHR/BP targeting vector, a 500-bp *Dra*III fragment of the 6.4-kb *Sac*I–*Eco*RI fragment was replaced by a 1.1-kb neomycin resistance gene (*neo*) cassette through blunt-end ligation (Fig. 1*A*). This replacement deleted the major portion of exon 4 and ≈500 bp of intron 4/5 of the GHR/BP gene. The *neo* cassette was flanked by mGHR/BP gene homologous sequences of 1.1 kb at the 5′ end and 4.8 kb at the 3′ terminus. A herpes simplex virus thymidine kinase gene (*tk*) cassette, which served as a negative selection marker (24), was placed at the 3′ end of the downstream mGHR/BP homologous sequence. Both the *neo* and *tk* cassettes were derived from plasmid pSSC9 (25).

Production of GHR/BP-Disrupted Mice. Embryonic stem (ES) cell transfection and selection were performed as described with minor modifications (26, 27). In brief, 10^7 E14 ES cells derived from 129/Ola mice (28) were transfected with 30–40 μg of linearized mGHR/BP targeting vector by electroporation, followed by positive-negative selection with G418 and ganciclovir (24). Colonies of ES cells with homologous recombination were identified by Southern blot analysis after *Bam*HI digestion of genomic DNA. The 1.7-kb *Bam*HI–*Sac*I fragment (P1) 5′ to the vector fragment and the *neo* cassette (P2) were labeled with ^{32}P by random priming and were used as hybridization probes (Fig. 1*A*). Heterozygous GHR/BP-

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disrupted (GHR/BP$^{+/-}$) ES cells were injected into BALB/c blastocysts. The injected blastocysts were transplanted into uteri of pseudopregnant BALB/c mice. Chimeric males were crossed to BALB/c females to produce GHR/BP$^{+/-}$ mice. The germ line transmission of GHR/BP$^{+/-}$ ES cells was monitored by detecting agouti-colored mice among the F1 offspring, and GHR/BP disruption was confirmed by Southern blotting. Homozygous GHR/BP-disrupted (GHR/BP$^{-/-}$) mice were produced by GHR/BP$^{+/-}$ inbreeding and confirmed by Southern blot analysis.

Western Blot Analysis of GHR Expression. Liver from GHR/BP$^{+/+}$, GHR/BP$^{+/-}$, and GHR/BP$^{-/-}$ mice was homogenized in lysis buffer as described (29). Total lysates (50 μg of protein) were fractionated on a 4–12% SDS/PAGE gel, transferred to Hybond membranes, and membranes probed with an antiserum directed against the mouse GHR intracellular domain (a gift from P. Frick and H. M. Goodman). The enhanced chemiluminescence (Amersham) system was used to visualize GHR protein.

GHBP Measurements. GHBP in mouse serum was measured by a standardized GH binding assay as described (30, 31). In brief, 50 μl of serum was incubated with 10^5 cpm [^{125}I]human (h)GH for 45 min at 37°C. (hGH binds with high affinity to the mouse GHBP and GHR.) Bound GH was separated from free GH by gel filtration and quantitated by peak integration. Nonspecific binding was determined in the presence of 2 μg/ml unlabeled hGH. Correction for the saturation by endogenous GH was made according to a saturation standard curve as described (30, 31).

GHR Determinations. GH binding to liver microsomal membranes was determined as described (29). Membrane aliquots (200 μg of protein) were incubated with [^{125}I]bovine (b)GH (50,000 cpm) at 4°C for 48 h. bGH, like hGH, binds to the mouse GHR. Here, bGH rather than hGH was used because the latter also binds to prolactin receptors that are present in liver membranes. (This is not a concern with serum, in which there is no detectable prolactin receptor/BP.) Nonspecific binding was determined in the presence of 200 nM of unlabeled bGH.

Hormone Measurements. Serum mouse (m)GH was measured by a polyclonal radioimmunoassay using reagents (mGH as tracer and standard, anti-rat GH antiserum) kindly provided by A. F. Parlow and the National Hormone and Pituitary Program. Serum IGF-I levels were determined after acid–ethanol extraction using a human IGF-I RIA kit as described (29). [Because mouse IGF-I differs from human IGF-I in 4 of 70 residues (32), the human assay underestimates rodent IGF-I by a factor of 2–3 (33).]

Statistical Analysis. Data were analyzed by contingency table, ANOVA, ANOVA for repeated measures, or Kruskal–Wallis ANOVA as appropriate, followed by the Newman–Keuls test. Data are given as mean ± SD. A *P* level of <0.05 was considered statistically significant.

RESULTS

Targeted Disruption of the GHR/BP Gene. Out of 512 clones dual-resistant to both G418 and ganciclovir, two contained the desired mutation in one allele of the GHR/BP gene. No additional integrations of the targeting vector were detected when analyzed by Southern blotting by using a *neo* hybridization probe (data not shown). Six chimeric animals were produced from both clones when microinjected into BALB/c blastocysts, and two chimeric males transmitted the disrupted allele to offspring, yielding GHR/BP$^{+/-}$ heterozygotes. Inbreeding of the F1 generation of GHR/BP$^{+/-}$ mice produced homozygous GHR/BP$^{-/-}$ mice.

The complete disruption of the GHR/BP gene was confirmed by Southern blotting (Fig. 1*B*). In homozygous animals, only the mutant band was detected. In addition, we have been unable to detect GHR/BP mRNA in livers of GHR/BP$^{-/-}$ animals even

by reverse transcription-PCR (data not shown). Genotyping of 243 offspring from GHR/BP$^{+/-}$ inbreeding revealed a frequency of 26% GHR/BP$^{+/+}$ (34 male, 28 female), 51% GHR/BP$^{+/-}$ (54 male, 70 female), and 23% GHR/BP$^{-/-}$ (33 male, 24 female), consistent with a Mendelian pattern of inheritance. The sex distribution was statistically not different among the three genotypes ($\chi^2 = 4.06$, $P > 0.1$).

Western blot analyses demonstrated no detectable GHR protein in the liver of GHR/BP$^{-/-}$ mice and decreased GHR levels in GHR/BP$^{+/-}$ animals (Fig. 1*C*). No GHBP was detected in sera of GHR/BP$^{-/-}$ mice (<30 pM) (Fig. 1*D*). Heterozygous GHR/BP$^{+/-}$ littermates had mildly decreased GHBP levels—a finding that did not reach statistical significance in this study. These findings are consistent with those in Laron syndrome individuals bearing mutations in the extracellular domain of the GHR gene (34, 35). As shown in Fig. 1*E*, the ability of liver from GHR/BP$^{-/-}$ mice to bind bGH was greatly decreased compared with GHR/BP$^{+/+}$ littermates although a small amount of residual binding was detectable. The residual binding is minimal but unexplained. Because bGH does not bind to prolactin receptors and because excess prolactin (1 μg/ml) did not diminish binding, the finding may indicate the presence of other type(s) of GH receptors. Alternatively, it may represent nonspecific background that is difficult to differentiate from zero binding. Heterozygous GHR/BP$^{+/-}$ mice had moderately decreased GH binding to liver, which differed statistically from both GHR/BP$^{+/+}$ and GHR/BP$^{-/-}$ animals.

Viability and Fertility of GHR/BP Knockout Mice. The average litter size from GHR/BP$^{+/-}$ inbreeding was 6.57 pups (Table 1). This is a typical litter size for GHR/BP$^{+/+}$ crosses at the same breeding age. The average age of first pregnancy for GHR/BP$^{-/-}$ females was delayed, implying delayed sexual maturation, as also has been reported in Laron syndrome patients (2). Inbreeding of GHR/BP$^{-/-}$ mice gave an average of 2.71 pups per litter, which is significantly smaller than those derived from GHR/BP$^{+/+}$ or GHR/BP$^{+/-}$ breeding. In addition, the perinatal or immediate postnatal mortality of newborns of GHR/BP$^{-/-}$ breeding was significantly higher than that of newborns of GHR/BP$^{+/-}$ or GHR/BP$^{+/+}$ breeding. The reason for smaller litter size and higher mortality of offspring from GHR/BP$^{-/-}$ breeding is not certain but may relate to placentation, "obstetrical problems" due to maternal–fetal size mismatch, and/or insufficient lactation to nourish the pups. Inspection of the stomach contents of neonates suggested decreased milk consumption in offspring from GHR/BP$^{-/-}$ parents compared with those of GHR/BP$^{+/-}$ parents.

Retarded Growth of GHR/BP Knockout Mice. At birth, no significant body size or weight differences were observed among GHR/BP$^{+/+}$, GHR/BP$^{+/-}$, and GHR/BP$^{-/-}$ littermates. At 3 weeks after birth, the GHR/BP$^{-/-}$ mice were significantly smaller than either GHR/BP$^{+/+}$ or GHR/BP$^{+/-}$ mice. With age, the difference in body size or weight increased progressively (Fig. 2*A–C*). Analysis of the growth curves showed a highly significant difference ($P < 10^{-6}$) for both sexes between GHR/BP$^{-/-}$ and either GHR/BP$^{+/-}$ or GHR/BP$^{+/+}$ animals. The growth curve up to 12 weeks for heterozygous males was indistinguishable from that of GHR/BP$^{+/+}$ males ($P \approx 0.35$) whereas for females the growth of heterozygotes was mildly but significantly retarded compared with GHR/BP$^{+/+}$ females ($P < 0.05$). At 8 weeks of age, body

Table 1. Viability and fertility of GHR/BP knockout mice

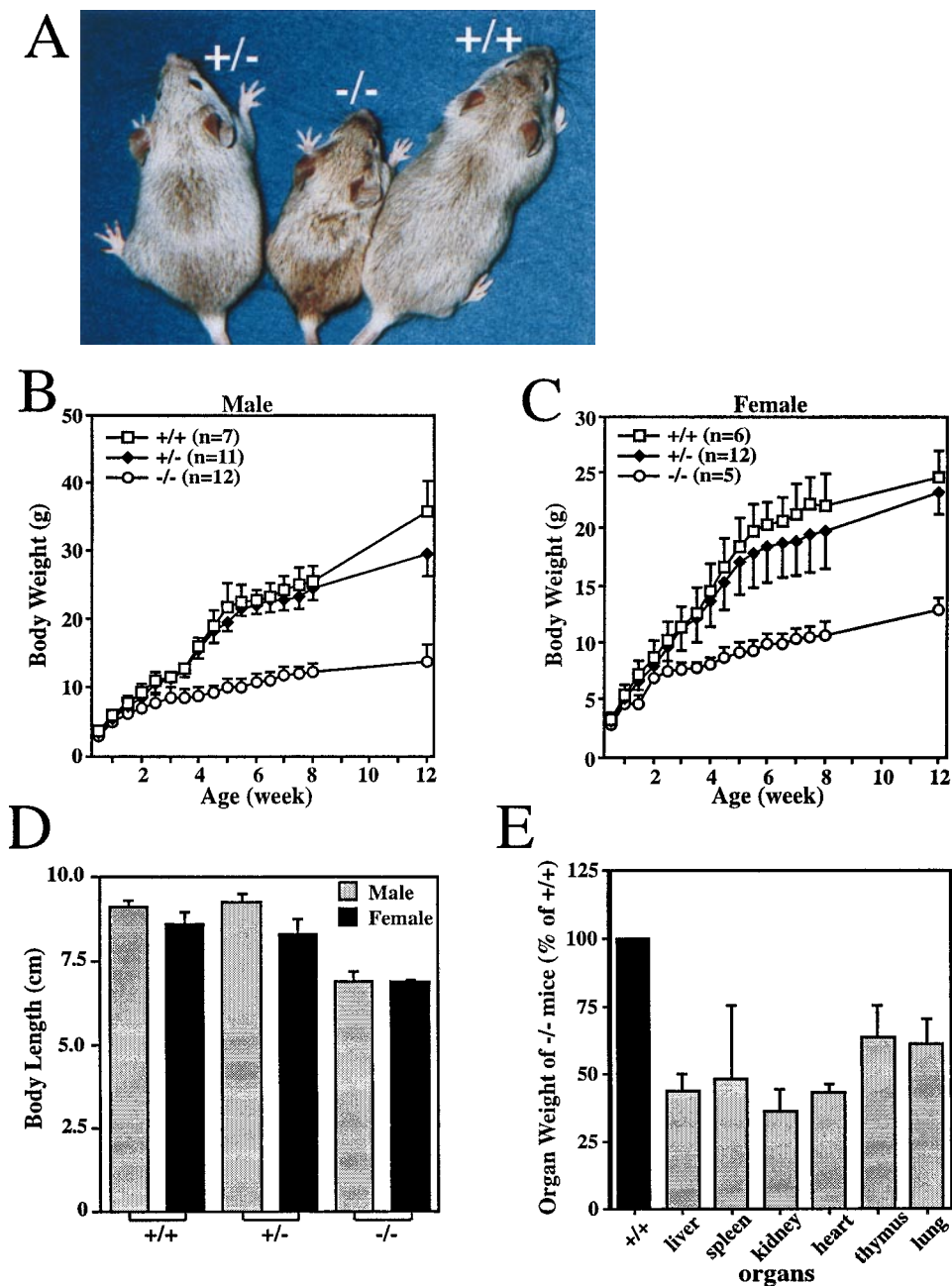| Cross	Litter size mean (range)	Perinatal mortality, %	Female sexual maturation, weeks
+/+ × +/+	6.90 pups (3–13)	5	6
+/− × +/−	6.57 pups (3–13)	6	6
−/− × −/−	2.71 pups (1–6)	26	10

FIG. 2.   Retarded somatic growth of GHR/BP-deficient mice. (*A*) Photograph of female GHR/BP$^{+/+}$, GHR/BP$^{+/-}$, and GHR/BP$^{-/-}$ mice at 4 weeks of age. (*B* and *C*) Growth curves of male and female mice. (*D*) Body length at 60 days of age, determined from the tip of the nose to the anus ($n = 3$ in each group). (*E*) Organ weights of 60-day-old GHR/BP$^{-/-}$ mice, expressed as a percentage of the corresponding mean organ weight in GHR/BP$^{+/+}$ mice ($n = 3$ for both groups).

length (nose–anus) in both sexes was significantly shorter in GHR/BP$^{-/-}$ than in GHR/BP$^{+/-}$ or GHR/BP$^{+/+}$ mice ($P < 0.01$) whereas heterozygous and GHR/BP$^{+/+}$ mice had similar body lengths. The length of 8-week-old GHR/BP$^{-/-}$ mice was significantly decreased when compared with GHR/BP$^{+/-}$ and GHR/BP$^{+/+}$ littermates (Fig. 2*D*). Female GHR/BP$^{+/-}$ mice were shorter than female GHR/BP$^{+/+}$ mice ($P < 0.05$), but the length difference between male GHR/BP$^{+/-}$ and GHR/BP$^{+/+}$ mice was not statistically significant. The weights of major organs of GHR/BP$^{-/-}$ mice also were significantly decreased ($P < 0.01$) compared with GHR/BP$^{+/+}$ or GHR/BP$^{+/-}$ littermates (Fig. 2*E*). Except for their smaller size, the organs exhibited no gross anatomic abnormalities.

Abnormal GH–IGF-I Axis of GHR/BP Knockout Mice. Serum GH levels for GHR/BP$^{-/-}$ mice were elevated greatly for both sexes compared with GHR/BP$^{+/+}$ and GHR/BP$^{+/-}$

mice ($P < 0.05$) (Fig. 3*A*). In contrast, the GH levels of GHR/BP$^{+/+}$ and GHR/BP$^{+/-}$ mice were comparable. The differences in GH levels between male and female mice were not statistically significant for any group. Serum IGF-I levels in GHR/BP$^{-/-}$ mice were decreased by ≈90% ($P < 0.001$) (Fig. 3*B*). IGF-I levels in GHR/BP$^{+/+}$ and GHR/BP$^{+/-}$ mice were not significantly different. These results are similar to those seen in human Laron syndrome and can be attributed to defective IGF-I generation in response to GH and lack of negative feedback of IGF-I on GH secretion.

Other Potential Abnormalities. No other abnormalities were evident in the homozygous or heterozygous knockout mice. They appear healthy and vigorous up to the date of this writing. In particular, their behavior was indistinguishable from that of their wild-type littermates. Lactation in GHR/BP$^{+/-}$ mice appeared adequate to feed their young.
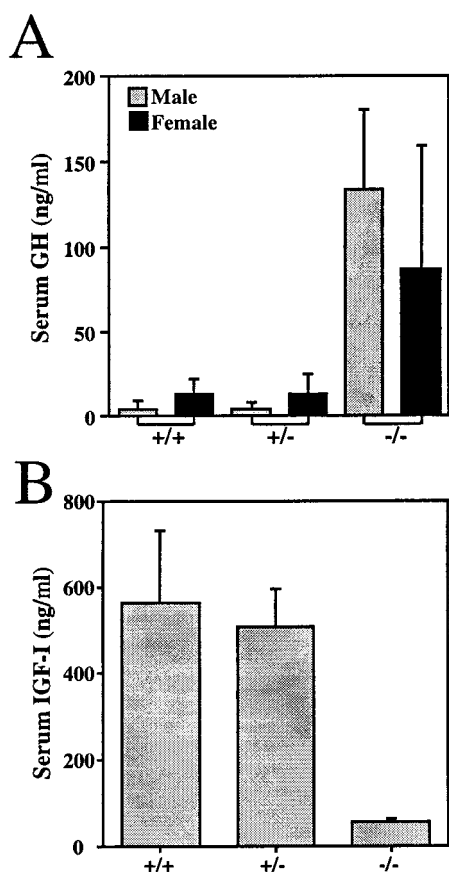
A

B

FIG. 3.　Serum GH (*A*) and IGF-I (*B*) concentrations of GHR/
BP$^{+/+}$, GHR/BP$^{+/-}$, and GHR/BP$^{-/-}$ mice. Blood was collected
from 3–4 mice of each genotype on day 30 and day 60 after birth.
Results from both time points were pooled.

DISCUSSION

The GHR/BP knockout mouse (Laron mouse) we generated
appeared to be a valid and suitable mammalian model for
human Laron syndrome. Its physical and biochemical pheno-
type greatly resembled the human disease. Salient features
shared between the two species are the absence of the GHR
and GHBP, the severe postnatal growth failure, delayed
puberty, and low IGF-I and elevated GH levels. The degree of
growth retardation is similar to that in the human syndrome.
It can thus be anticipated that this model will be useful in
providing new answers to a number of unresolved questions in
this field. Examples are: the long term growth response and
final height/size achievement with IGF-I therapy; the effect of
GHR/BP deficiency on longevity, immune function, gonadal
function, fertility, and fecundity; as well as its impact on
pathological processes suspected to be linked to GH (e.g.,
diabetic microvascular disease). The homogeneous genetic
background, rapid breeding, short growth phase, and short
life-span of this mouse model will accelerate the acquisition of
knowledge that may take decades to accumulate in human
Laron syndrome. Furthermore, the model will permit a de-
tailed study of the absence of the GHR and GHBP on body
composition and tissue characteristics at the histological, his-
tochemical, and molecular levels—information that cannot be
obtained in humans for ethical and technical reasons. Finally,
the Laron mouse will serve as a useful null background for
studies on the impact of gene replacement (e.g., GHBP vs.
GHR vs. both) in a global or tissue-specific manner for
physiological studies of the turnover and metabolic fate of
GHBP and for dissecting the relative contributions of endo-

crine vs. paracrine IGF-I to growth and development through
systemic IGF-I administration.

Some new insights already have been gained at this early
stage. The birth weight of GHR/BP-deficient mice born of
heterozygous parents was identical to that of wild-type mice.
The role of GH in fetal growth and development, albeit minor,
has been a matter of controversy because both GHR and GH
are expressed in the fetus (36, 37). Our present data do not
support a role of fetal GH in intrauterine growth up to the
relatively "immature" stage at which mice are born. Inspection
of the growth curves indicates that the GH dependence of
postnatal growth commences at or shortly after birth rather
than after a delay, as is sometimes suggested. There is a GHR
gene dosage effect on the biochemical phenotype in that
heterozygotes have GHR and GHBP levels intermediate be-
tween wild-type and homozygous knockout animals. The
effect of heterozygosity on the physical phenotype is minimal
but discernible within the first 12 weeks; growth in females
appears more affected by gene dosage than in males. Visceral
organs of Laron mice are small in size, with no other gross
abnormality. The male/female ratio of homozygous GHR/BP
knockout mice (1.37) is indistinguishable from that of wild-
type mice (1.21), as would be expected for an autosomally
transmitted disorder. This is a reassuring observation in view
of the striking but unexplained female preponderance of
Laron syndrome in the Ecuadorian population of Loja (2, 38).
The normal sex ratio of Laron mice is concordant with the
worldwide male/female ratio for Laron syndrome, which is
close to unity (2, 8, 10); it suggests that deviations from unity
in specific populations are due to either chance or factors
independent of the GHR.

As mentioned above, an avian model for GHR deficiency
exists in the form of the genetically heterogeneous sex-linked
dwarf chicken (11). Although that model shares some features
with mammalian GHR/BP deficiency, it is not an ideal model
for human Laron syndrome for several reasons. Taxonomic
and anatomic differences between mammals and birds render
direct comparisons difficult. IGF regulation and IGF action
may be different in birds and mammals (39, 40). The repro-
ductive and perinatal biology of birds deviates greatly from
that in mammals.

The growth curves of our Laron mice are very similar to that
of the little (*lit/lit*) mouse that has isolated GH deficiency (41).
In contrast, the Snell and Ames dwarf mice are more growth-
retarded, presumably because they are hypothyroid in addition
to being GH-deficient (42, 43).

Fertility appears to be preserved in homozygous GHR/BP
knockout mice of both genders, indicating that GH action is
not essential for fertility. This is consistent with several re-
ported instances of fertility in both men and women affected
with Laron syndrome. However, for mice born of two GHR/
BP$^{-/-}$ parents, fetal outcome is not normal in that they have
fewer siblings and decreased perinatal survival. It is presently
not clear what is responsible for this phenomenon, but fetal–
maternal size mismatch *in utero* and during birth or abnormal
lactation or suckling are potential explanations. No corre-
sponding information exists in humans because, to our knowl-
edge, no union of two patients affected with Laron syndrome
has been reported.

The residual GH binding in livers (and kidneys; not shown)
of homozygous knockout mice is of interest. Although we did
not detect GHR mRNA, we consistently found a small amount
of GH binding activity above background. This may suggest
that another GHR exists, a notion that has been postulated
(22). If extant, this putative GHR does not appear to contrib-
ute to somatic growth as the animals are dwarfed, but it is an
intriguing possibility that it may be involved in other GH-
specific activities, such as metabolic and/or diabetogenic
action. Further work will have to be performed to determine
whether the residual GH binding activity represents a receptor.

The Laron mouse will perhaps be most useful in two areas of long term development in which human data are still limited or lacking. One is aging and longevity. It recently has been reported that Ames dwarf mice live significantly longer than their normal siblings, and GH deficiency has been implicated in this process (44). The life-span of GHR/BP knockout mice may yield new insights into a possible role of GH and IGF-I in senescence. Second, the Laron mouse may help answer a long-standing question in this field, namely whether IGF-I can promote long term linear growth and normal adult size in the absence of GH action. Isaksson *et al.* (45) have proposed that epiphyseal prechondrocytes must first differentiate under the influence of GH and only when differentiated undergo IGF-I-mediated clonal expansion, which leads to linear bone growth. The differentiated chondrocyte pool is limited and cannot be replenished by recruiting new chondrocytes except through continued GH action. Hence, they have suggested that IGF-I therapy of Laron syndrome may be effective for only a limited time, namely until the differentiated chondrocyte pool is depleted. Trials with chronic IGF-I therapy of Laron patients have been going on for ≈3–4 years, and IGF-I effectiveness is indeed declining with time (46, 47). The same is true, however, for GH therapy in GH deficiency, although perhaps to a lesser degree. It will take many years to determine the long term efficacy of IGF-I in human Laron syndrome. The same experiment, performed in the Laron mouse, will yield an answer regarding final size in 6 months. Thus, we hope that the present GHR/BP knockout mouse will provide the keystone in the proof of the somatomedin hypothesis.

In summary, we report the generation and initial data on the dwarf phenotype of a mouse with a GHR/BP gene knockout (the Laron mouse). In its homozygous and heterozygous form, the Laron mouse exhibits a phenotype that greatly resembles the human genetic disease known as Laron syndrome. Therefore, this GHR/BP-deficient mouse will be a useful model in the study of many unresolved aspects of GH-IGF-I function, such as aging, longevity, tumorigenesis, and diabetes and its complications.

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