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(Epi)genotype–phenotype correlations in Beckwith–Wiedemann syndrome

Alessandro Mussa, Silvia Russo, [...], and Giovanni Battista Ferrero

Abstract

Beckwith–Wiedemann syndrome (BWS) is characterized by cancer predisposition, overgrowth and highly variable association of macroglossia, abdominal wall defects, nephrourological anomalies, nevus flammeus, ear malformations, hypoglycemia, hemihyperplasia, and organomegaly. BWS molecular defects, causing alteration of expression or activity of the genes regulated by two imprinting centres (IC) in the 11p15 chromosomal region, are also heterogeneous. In this paper we define (epi)genotype–phenotype correlations in molecularly confirmed BWS patients. The characteristics of 318 BWS patients with proven molecular defect were compared among the main four molecular subclasses: IC2 loss of methylation (IC2-LoM, $n=190$), IC1 gain of methylation (IC1-GoM, $n=31$), chromosome 11p15 paternal uniparental disomy (UPD, $n=87$), and cyclin-dependent kinase inhibitor 1C gene (*CDKN1C*) variants ($n=10$). A characteristic growth pattern was found in each group; neonatal macrosomia was almost constant in IC1-GoM, postnatal overgrowth in IC2-LoM, and hemihyperplasia more common in UPD ($P<0.001$). Exomphalos was more common in IC2/*CDKN1C* patients ($P<0.001$). Renal defects were typical of UPD/IC1 patients, urethral malformations of IC1-GoM cases ($P<0.001$). Ear anomalies and nevus flammeus were associated with IC2/*CDKN1C* genotype ($P<0.001$). Macroglossia was less common among UPD patients ($P<0.001$). Wilms' tumor was associated with IC1-GoM or UPD and never observed in IC2-LoM patients ($P<0.001$). Hepatoblastoma occurred only in UPD cases. Cancer risk was lower in IC2/*CDKN1C*, intermediate in UPD, and very high in IC1 cases ($P=0.009$). In conclusion, (epi)genotype–phenotype correlations define four different phenotypic BWS profiles with some degree of clinical overlap. These observations impact clinical care allowing to move toward (epi) genotype-based follow-up and cancer screening.

Introduction

Beckwith–Wiedemann syndrome (BWS) (OMIM #130650) is the commonest genetic overgrowth condition, with a prevalence approximating 1 in 10 000 live births.¹ BWS has a wide clinical spectrum including several variably associated anomalies: its cardinal features, beside overgrowth, include abdominal wall defects, macroglossia, nephrourologic malformations, hemihyperplasia, hyperinsulinemic hypoglycemic, ear anomalies (lobe creases or helical pits), hemangiomas and nevus flammeus at the glabella, and organomegaly.² The diagnosis can be established clinically by these diagnostic criteria, although none is mandatory.³ BWS is a cancer predisposition syndrome; malignancy risk is estimated to range between 5 and 15%, being highest at birth and approaching the baseline of the general population before puberty onset.⁴ The tumor spectrum mostly comprises embryonal histotypes, with Wilms' tumor, hepatoblastoma, and adrenocarcinoma being the most frequent ones.

The variability of BWS clinical spectrum is paralleled by comparable (epi)genetic heterogeneity at the molecular level.^{2, 5, 6} BWS is a paradigm of disorders associated with defective genomic imprinting, a process consisting in a parent-of-origin-specific gene expression. BWS is caused by altered expression of two gene clusters involved in cell cycle progression and somatic growth control regulated by two independent imprinting centres (IC1 and IC2) at chromosome 11p15.5. IC1 and IC2 are characterized by differential methylation of their maternal and paternal alleles. Different molecular mechanisms lead to unbalanced expression of the imprinted genes in BWS; ~50% of cases are caused by loss of methylation at IC2 (IC2-LoM), resulting in reduced expression of cyclin-dependent kinase inhibitor 1C gene (*CDKN1C*), normally expressed by the maternal chromosome only. Maternal *CDKN1C* loss-of-function variants also account for 5–10% of cases and are responsible for half of the inheritable ones. From 5 to 10% of BWS cases are caused by gain of methylation at IC1 (IC1-GoM), which results in biallelic expression of the insulin growth factor 2 gene – normally expressed by the paternal allele – and reduced expression of the oncosuppressor *H19* gene – normally expressed by the maternal allele. Recently, it has been demonstrated that 20% of such cases are caused by inheritable *OCT4/SOX2* binding site cis genetic defects.^{7, 8} Altered expression at both gene clusters is observed in cases with mosaic paternal uniparental disomy (UPD) (20%: genome-wide UPD is growingly found in a subset of UPD cases and associated with additional phenotypic features. Overall, <1% of BWS cases are caused by chromosomal rearrangements such as duplications, translocations, inversions, deletions, involving genes into the IC gene clusters. Finally, ~15% of clinically diagnosed cases have no detectable molecular defect in spite of a clear-cut phenotype.^{2, 5, 6}

Although recent investigations proved the association between molecular alterations, clinical features, and cancer risk,^{4, 9, 10, 11, 12, 13} the complex (epi)genotype–phenotype relationship in BWS has still to be fully unraveled. Here, we report the clinical and molecular characterization of a large cohort of BWS patients that allows detailed (epi)genotype–phenotype correlations and supports the hypothesis that different (epi)genetic alterations are associated with specific phenotypes in BWS.

Materials and methods

Phenotyping

Overall, 318 patients were ascertained via the Italian National BWS Network following referral to the laboratories providing genetic testing for BWS in Italy (Laboratory of Cytogenetics and Molecular Genetics, Istituto Auxologico Italiano, Milan and DiSTABiF, Second University of Naples, Italy). Through the involvement of the major clinical genetics centers, clinical information was collected by the physicians who made the diagnosis, requested the genetic testing, and followed-up of cases. Using a standardized questionnaire, physicians were asked to specify the presence/absence of the features of BWS and provide informations relevant to phenotype and tumor development. Macrosomia was defined as birth weight >90th percentile according to gestational age.¹⁴ Discrete BWS features (eg, macroglossia, hemihyperplasia, nevus flammeus) were diagnosed by evaluation by respective specialists (eg, odontostomatologist, ortopedics, dermatologist). Data were further implemented through a search in the AIEOP (Italian Onco-Hematological Association) tumor registry. Therefore, tumor occurrence is updated to the latest available visit and double checked via a tumor registry allowing a more precise definition of the tumor risk during the follow-up. Patients with at least two BWS criteria (among abdominal wall defects presence and severity, macroglossia, macrosomia, embryonal tumor, ear malformations, organ enlargement, nevus flammeus, hemihyperplasia, nephrourological malformations, cleft palate, hypoglycemia, family history of BWS, polyhydramnios) and proven molecular diagnosis were included. Four cases with isolated hemihyperplasia and positive molecular tests were also included. To provide a fully meaningful analysis of the correlation between phenotype and (epi)genotype, negative cases were not taken into consideration to avoid ascertainment bias owing to overlapping conditions.

Genotyping

All patients or the parents provided written informed consent to the genetic testing. DNA was extracted from peripheral blood lymphocytes. Methylation analysis of the 11p15.5 chromosomal regions containing IC1 and IC2 was carried out in all patients and performed either by Southern blotting ($n=170$), COBRA ($n=45$)¹⁵ or Methylation-Sensitive Multiple Ligation Probe Amplification (MS-MLPA MRC-HOLLAND kit) ($n=103$).¹⁶ The results obtained by these techniques have been shown to be comparable.^{16, 17} In patients with suspected UPD, confirmation was obtained by microsatellite analysis of probands and parents, as described.¹⁸ The presence of genome-wide UPD was tested in 28 UPD patients by microsatellite analysis and single-nucleotide polymorphism array. *CDKN1C* gene sequencing as described elsewhere¹⁹ was carried out in 154 patients selected on the basis of negativity of methylation sensitive tests plus 2 of the above-mentioned BWS diagnostic criteria and either familiarity for BWS or signs/malformations highly specific for *CDKN1C* variants (as palatoschisis or omphalocele).³ Pathogenicity prediction of *CDKN1C* variants was tested by the bioinformatic tools PolyPhen-2 (Polymorphism Phenotyping), SIFT (Sorting Intolerant From Tolerant), and PROVEAN (Protein Variation Effect Analyzer). Variants were submitted to LOVD (Leiden Open Variation Database 3.0, www.lovd.nl, variants #0000058604, #0000058622, #000005860, #000005862, #0000058601, #0000058602, #0000055971, #0000055979, #0000055977, #0000055899, submitter ID 01227).²⁰

Statistical analysis

Data were summarized with descriptive statistics. Comparisons among the molecular groups was conducted by 2×2 (for each category, versus all other categories) or comparing categories by 3×2 or 4×2 Fisher's exact tests or, in case of expected frequencies ≥ 5 , χ^2 -test with Yates correction, as appropriate. Two-tailed P -values <0.05 were considered as significant. Data were analyzed by SPSS 13.0 (IBM Software, Armonk, NY, USA) and Prism GraphPad 5.0 (GraphPad Software, La Jolla, CA, USA).

Results

A total of 318 patients with confirmed epimutation in 11p15.5 or *CDKN1C* variant were characterized. The following molecular anomalies were identified: 190 IC2-LoM (184 epigenetic anomalies, 5 already published cases with familial IC2 duplications²¹ and 1 IC2 deletion), 87 UPD carriers, 31 IC1-GoM (21 already published cases^{22, 23} including one IC1 duplication, one translocation, 11 familial microdeletions^{24, 25, 26}), 10 *CDKN1C* variants (all unrelated cases, 9 maternally inherited). None of the patients tested was positive for genome-wide UPD. The four cases with isolated hemihyperplasia were affected by UPD ($n=2$) or IC2-LoM ($n=2$).

The prevalence of the BWS features in the four subgroups is summarized in Figure 1. The growth patterns showed relevant differences across the molecular subtypes (Figure 1a and d). In patients with IC1-GoM, neonatal macrosomia was almost constant and much more common than in the other subgroups ($P=0.002$). The prevalence of postnatal overgrowth showed minor differences, being slightly higher in patients with IC2-LoM ($P=0.016$) and *CDKN1C* variants and lower in those with UPD ($P=0.049$). The latter group had an incidence of hemihyperplasia of almost twofold that of IC2-LoM/IC1-GoM patients ($P<0.001$), whereas hemihyperplasia was not observed at all in *CDKN1C* variants ($P<0.001$). Also the distribution of the severity of abdominal wall defects varied extensively among BWS subtypes (Figure 1e and h, Figure 2). Their prevalence was higher in the IC1-GoM group ($P<0.001$, 70% of cases), in which the defects were mostly minor ($P<0.001$) with diastasis recti prevailing ($P=0.007$). Minor defects were also common among UPD patients, but with an overall prevalence of abdominal wall defects much lower than in other groups (48.3%, $P<0.001$). Patients with IC2-LoM had an intermediate prevalence of abdominal wall defects (66.8%) and showed an increased risk of major ones (omphalocele 30.0%, $P<0.001$). Patients with *CDKN1C* variants showed very high incidence of omphalocele (70%, $P=0.001$). Macroglossia was present in most of the cases with IC1-GoM (90.3%) and IC2-LoM (88.4%), but was less common in UPD (69.0%) and *CDKN1C* variant cases (70%) ($P<0.001$) (Figure 1i). Ear signs were more represented among IC2-LoM and *CDKN1C* variant patients (50.5% and 60%, respectively) than among IC1-GoM or UPD cases (22.6% and 39.1%, respectively, $P=0.013$) (Figure 1j). Similar differences

were observed for the occurrence of nevus flammeus (48.4%, 50.5%, 22.6%, and 34.5%, respectively, $P=0.016$) (Figure 1k). Cleft palate was more common in *CDKN1C* variant patients (Figure 1l), but not significantly. Organ enlargement was reported in 67.7% of IC1-GoM cases, significantly higher than the occurrence in IC2-LoM (27.9%), UPD (36.8%), and *CDKN1C* variant (10%) cases ($P<0.001$) (Figure 1m). Kidney abnormalities were more frequently detected in IC1-GoM (32.5%) and UPD (26.4%) patients, as compared with IC2-LoM (8.9%) and *CDKN1C* (20%) variant cases ($P<0.001$) (Figure 1n). Ureteral malformations prevalence was highest among IC1-GoM cases (22.6%, $P<0.001$) and lower in other subtypes (overall 5.2%) (Figure 1o). Fourteen (4.4%) patients conceived with the use of assisted reproduction techniques: 10 cases had IC2-LoM and 4 had UPD. Preterm birth (<37 weeks of gestation) was more common in cases with *CDKN1C* variants (71.4%) and IC2-LoM cases (41.3%, $P<0.001$) than in other molecular subtypes (UPD 18.1%, IC1-GoM 28.6%) (Figure 1r). Polyhydramnios was more common among IC1-GoM patients (35.5%, $P=0.016$) than IC2-LoM (15.3%), UPD (14.9%) or *CDKN1C* variant (0%) cases (Figure 1s). We observed no difference in the occurrence of hypoglycemic (Figure 1t). Three patients deceased (1 IC2-LoM, 1 UPD, 1 IC1-GoM) of prematurity-related complications (two cases of sepsis consequent to urinary tract infection owing to ureteral malformations, one of respiratory insufficiency). Concerning cancer occurrence, 33 patients developed a neoplasm during their follow-up, which lasted on average 9.8 ± 7.3 (median 8.9) years (age range 0–2 years $n=67$, 2–4 years $n=56$, 4–8 years $n=75$, >8 years $n=120$). Twenty-four malignant neoplasms were reported in 23 patients (7.2%) (Figure 1p) and 14 benign tumors (Figure 1q) were observed in 14 cases of which 3 also had a malignancy. No tumor was recorded in *CDKN1C* variant patients, whereas the incidence of malignant neoplasms varied significantly in the other three subgroups: 2.1% in IC2-LoM, 14.9% in UPD, and 25.8% in IC1-GoM patients ($P<0.001$). Wilms' tumor developed only in patients with IC1-GoM or UPD, being clearly the prevalent cancer in IC1-GoM patients ($P<0.001$) (Tab. 1). Hepatoblastoma was the most common tumor among UPD patients and was not reported in the other molecular subgroups ($P=0.003$). The tumor-free probability curves according to the molecular defects are depicted in Figure 3. Age at tumor diagnosis in IC1-GoM, UPD, and IC2-LoM patients was 13.8 ± 9.3 , 19.1 ± 18.6 , and 13.6 ± 3.2 months, respectively. Mean age at the diagnosis for Wilms' tumor, hepatoblastoma, and neuroblastoma was 18.6 ± 13.0 , 16.2 ± 26.9 , and 16.0 ± 8.2 months. There was a significant difference in the incidence of benign tumors ($P=0.009$), which were increasingly more common in IC2-LoM, UPD, and IC1-GoM patients (Figure 1s). The histotypes seen in the molecular subgroups are reported in Table 1.

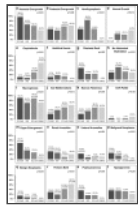


Figure 1

Differences in the prevalence of the features in the molecular subtypes of the syndrome: (a) neonatal overgrowth, (b) postnatal overgrowth, (c) hemihyperplasia, (d) normal growth, (e) omphalocele, (f) umbilical hernia, (g) diastasis recti, (h) no abdominal ...

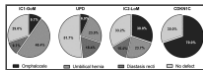


Figure 2

Enrichment of the abdominal wall defects in the molecular groups.

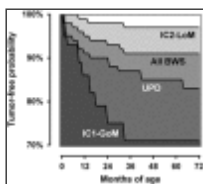


Figure 3

Kaplan–Meier plot of the tumor-free interval (malignant neoplasms only) in the three main molecular subtypes of Beckwith–Wiedemann syndrome (BWS). IC2-LoM: imprinting center 2 loss of methylation, UPD: paternal uniparental disomy; IC1-GoM:

Neoplasm	Molecular subtype				P-value
	IC2-LoM	UPD	IC1-GoM	CDKN1C	
Neoplasms	2 (2.1%)	4 (14.9%)	10 (25.8%)	0	<0.001
Malignant	0	1	1	0	<0.001
Benign	2	3	9	0	0.009
Hepatoblastoma	0	1	0	0	0.003
Wilms' tumor	0	0	1	0	0.001
Neuroblastoma	0	0	1	0	0.001
Other	2	2	8	0	0.009
Embryonal	0	0	1	0	0.001
Stromal	0	0	1	0	0.001
Other	2	2	8	0	0.009

Table 1

Summary of the neoplasms reported

Correlations between each of the BWS features were explored in the cohort. We found significant association between malignant neoplasms and hemihyperplasia ($P<0.001$) and organ enlargement ($P=0.030$), Wilms' tumor and hemihyperplasia ($P=0.024$), hepatoblastoma and hemihyperplasia ($P=0.019$), polyhydramnios and ureteral anomalies ($P=0.017$), nevus flammeus and ear malformations ($P<0.001$), organomegaly and abdominal wall defects ($P=0.038$), umbilical hernia ($P=0.039$), and diastasis recti ($P=0.018$). Fourteen among Wilms' tumor, hepatoblastoma, and pancreatoblastoma cases occurred in enlarged organs.

Discussion

BWS is characterized by one of the widest phenotypic spectra of syndromic developmental disorders, ranging from lethal to mild and incomplete forms. This highly variable phenotypic expression is paralleled at the molecular level by a complex heterogeneity of (epi)genetic defects at chromosome 11p15.5. Correlations between genotype and phenotype have been previously reported in other BWS cohorts.^{4, 9, 10, 11, 13, 27, 28, 29} In particular, omphalocele, ear signs, and nevus flammeus were associated with IC2 LoM or *CDKN1C* variants, hemihyperplasia with UPD, and Wilms' tumor with IC1-GoM or UPD.^{4, 9, 10, 11, 13, 27, 28, 29} In this study we further investigated these correlations providing data on a large cohort of fully characterized BWS patients with 11p15 region molecular defects. Our analysis evidences in the four BWS molecular subtypes differences in the incidence of many phenotypic traits, such as growth pattern, prevalence and severity of abdominal wall defects, macrosomia, nevus flammeus, ear signs, renal malformations, ureteral anomalies, organ enlargement, polyhydramnios, cancer incidence, and histotypes.

This analysis allows to define phenotypic profiles that are characteristic of the different molecular subgroups. Patients with IC1-GoM are constantly macrosomic at birth and commonly present abdominal wall defects – usually minor – consistent with organ enlargement; approximately one-third has renal anomalies and ureteral malformations that correlate with higher occurrence of polyhydramnios. IC2-LoM patients show an excess of premature births. In contrast, neonatal macrosomia is much less represented in this group and they rather present postnatal overgrowth. It is important to underline that the prevalence of macrosomia in BWS cohorts depends on its definition; we opted for the permissive definition of neonatal weight >90th percentile as of more diffuse usage, already employed in the definition of BWS diagnostic criteria,⁴ and used to define the large-for-gestational-age newborn in our setting.¹⁴ We confirm the increased prevalence of omphalocele in IC2-LoM further show that they have lower incidence of organ enlargement, suggesting that wall defects are primarily caused by developmental anomalies of the abdominal wall rather than consequent to increased abdominal pressure; nevus flammeus and ear signs are also particularly frequent in IC2-LoM patients (about half of the cases).

As previously reported, UPD patients typically present with hemihyperplasia; most of them have no abdominal wall defect, the others usually display only minor ones; concerning the other BWS-associated features, they generally show an intermediate prevalence with respect to IC1-GoM and IC2-LoM, consistent with the extent of the molecular defect, which affects both domains of the 11p15.5 cluster. It is worth to mention that there are conflicting results concerning the existence of a correlation between phenotype severity in UPD cases and the level of somatic mosaicism or the extent of the chromosomal isodisomy.^{30, 31, 32, 33} In this study, however, we evaluated only the presence and not the severity of the single BWS features and did not explore the two above-mentioned molecular factors. Moreover, we excluded only a fraction of the UPD patients genome-wide UPD, a genetic phenomenon linked to a further increase in cancer risk³⁴ and additional phenotypic features.³⁵

As concerns *CDKN1C* patients, a striking overlap with IC2-LoM phenotype was evident; they shared a similar growth pattern with low incidence of neonatal macrosomia and frequent occurrence of postnatal overgrowth, excess of preterm births, comparable proportion of ear signs and nevus flammeus, low prevalence of organ enlargement. Consistent with previous observations,^{9, 10, 19, 29} *CDKN1C* patients were characterized by the highest prevalence of omphalocele and cleft palate. Moreover, we did not detect any case of hemihyperplasia in this group. However, conclusions on *CDKN1C* phenotype should be drawn cautiously given the small number of patients included. Moreover, as we sequenced *CDKN1C* gene in a subset of selected patients, our data are prone to be biased.

It is well known that BWS is more common among patients conceived by artificial reproduction technique;^{36, 37, 38} we encountered a 4.4% prevalence of this phenomenon, confirming data from previous reports and showing a higher prevalence than that reported in the Italian population (1.7%).³⁹

As concerns tumor risk, the overall prevalence of cancer approximates 8%, consistent with other studies.¹⁰ It is well established that patients with telomeric defects (IC1-GoM/UPD) have a major risk of tumors, especially Wilms' tumor, whereas patients with defects of the centromeric domain (IC2-LoM/*CDKN1C* variant) have a lower risk.^{4, 9, 10, 40} Our data also point to a gradient of oncogenic risk between the three main molecular subgroups. At one end of the spectrum, patients with IC2-LoM have a very low risk of tumors (<2%) and do not develop Wilms' tumors. At the other end of the spectrum, patients with IC1-GoM have a very high tumor risk (25%) and are particularly prone to Wilms' tumor development. Between the two groups, UPD patients show an intermediate oncogenic risk (15%) and can develop histotypes seen in both IC1-GoM and IC2-LoM cases; furthermore, UPD cases show a previously unreported predisposition to hepatoblastoma, the second more common histotype of BWS, occurring in 1.6% of BWS patients, that is, 6% of UPD cases. We did not observe hepatoblastoma in the other molecular subgroups, but cannot conclude that hepatoblastoma occurs only in UPD cases, as three cases have been described in IC2-LoM patients previously.^{4, 10}

Few data are available on benign neoplasms in BWS;^{40, 41, 42} interestingly, their incidence is a gradient across the molecular subtypes paralleling that of malignancies: highest (~13%) in IC1-GoM, intermediate (~7%) in UPD, and lower (~4%) in IC2-LoM patients. Among benign histotypes observed, hepatic angiomas were prevailing, and no differences were detectable across the molecular subgroups.

Several of the correlations evidenced between (epi)genotype and phenotype consolidate previous observations (Table 2).^{4, 9, 10, 11, 13, 27, 28, 29} Some aspects emerge as new: in particular, the significant association between hepatoblastoma and UPD may have relevant implications for cancer screening, the association between IC2-GoM and ureteral defects and polyhydramnios may have implications for the neonatal nephrourological management, the higher incidence of benign neoplasm paralleling the distribution of the malignant ones should be taken into considerations during patients' follow-up. Finally, IC2-LoM/*CDKN1C* variant patients display a higher rate of postnatal overgrowth, poorly

studied before; as in these molecular subgroups neonatal macrosomia is rarer than in UPD/IC1-GoM ones¹³ clinicians should be aware that these molecular subtypes of BWS may display specific growth patterns after the neonatal period.

Table 2
Significant (epi)genotype-phenotype correlations in the three most recent correlation studies on BWS

Table 2

Significant genotype–phenotype correlations in the three most largest recent correlation studies on BWS

Based on these and recent findings,^{4, 13} we suggest a revision of the guidelines for tumor surveillance that takes into consideration the molecular defects. At present, cancer surveillance programs for BWS patients are based on a 3–6 months abdominal ultrasound up to 7–8 years of age to detect Wilms' tumor and 2–3 months serum alpha-fetoprotein determinations up to 4 years of age to screen for hepatoblastoma.⁴ Cost-effectiveness of ultrasound screening is proven;⁴³ we hypothesize that patients with IC1-GoM may benefit from an intensification of abdominal ultrasound during the first 3 years of life, as most of Wilms' tumors are diagnosed before that age and appears justified by their 25% chance of developing a Wilms' tumor, the well-proven beneficial effect of early diagnosis, and the low invasivity of abdominal ultrasound.⁴⁴ Conversely, the dosage of the tumor marker serum alpha-fetoprotein as a screening method for the early diagnosis of hepatoblastoma is debated, given the complexity of its interpretation in childhood,^{45, 46} the low incidence of hepatoblastoma and the invasivity of frequent blood draws, which is commonly responsible for the lack of adherence to screening protocols.⁴⁷ We believe that monitoring is worthwhile at least in UPD patients, given their high risk of hepatoblastoma. More questionable is the employment of ultrasound and tumor markers screening in IC2-LoM cases, given the low risk and the occurrence of histotypes for which the advantage of these screening methods is still unproven. In this molecular group, which represents >50% of BWS cases, clinical research should be focused in assessing its cost-effectiveness and further studies are needed to assess its ability to detect IC2-LoM-related malignancies and the actual impact of the detection timing on their management and treatment. A clinical follow-up with *ad hoc* instrumental/laboratory investigations may prove to be a reasonable alternative.

Conclusion

In conclusion, although none of the BWS phenotypes can be considered specific to a molecular anomaly, the relevant differences observed in the four molecular subtypes allow to speculate that BWS could be separated into four different conditions with different malformative pattern and specific phenotypic profile despite some degree of clinical overlap. This composite view of this syndrome likely has relevant implications and does impact on clinical care of patients allowing to move toward a (epi)genotype-based follow-up.

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Appendix

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Notes

The authors declare no conflict of interest.

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