Silver-Russell syndrome: a dissection of the genetic aetiology and candidate chromosomal regions

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Abstract

The main features of Silver-Russell syndrome (SRS) are pre- and postnatal growth restriction and a characteristic small, triangular face. SRS is also accompanied by other dysmorphic features including fifth finger clinodactyly and skeletal asymmetry. The disorder is clinically and genetically heterogeneous, and various modes of inheritance and abnormalities involving chromosomes 7, 8, 15, 17, and 18 have been associated with SRS and SRS-like cases. However, only chromosomes 7 and 17 have been consistently implicated in patients with a strict clinical diagnosis of SRS. Two cases of balanced translocations with breakpoints in 17q23.3-q25 and two cases with a hemizygous deletion of the chorionic somatomammotropin gene (CSH1) on 17q24.1 have been associated with SRS, strongly implicating this region. Maternal uniparental disomy for chromosome 7 (mUPD(7)) occurs in up to 10% of SRS patients, with disruption of genomic imprinting underlying the disease status in these cases. Recently, two SRS patients with a maternal duplication of 7p11.2-p13, and a single proband with segmental mUPD for the region 7q31-pter, were described. These key patients define two separate candidate regions for SRS on both the p and q arms of chromosome 7. Both the 7p11.2-p13 and 7q31-pter regions are subject to genomic imprinting and the homologous regions in the mouse are associated with imprinted growth phenotypes. This review provides an overview of the genetics of SRS, and focuses on the newly defined candidate regions on chromosome 7. The analyses of imprinted candidate genes within 7p11.2-p13 and 7q31-qter, and gene candidates on distal 17q, are discussed.

Keywords: Silver-Russell syndrome; imprinting; mUPD(7); candidates

Table 1 Clinical features and their reported frequencies in SRS, and genetic subgroups of SRS. The percentage of SRS features in two separate groups of SRS patients are listed.14 These features could not be combined as certain clinical conditions were not examined consistently in both sets of patients. These serve as a reference for the frequency of features observed in specific genetic categories of SRS patients mentioned in the text. Where the patient number is small the frequencies are given as fractions, not percentages. Traits either not reported or not evaluated are blank. Non-SRS features apparent in the SRS-like patients with abnormalities of chromosomes 18 and 8 are not included, but discussed in the text.

<table>
<thead>
<tr>
<th>Clinical feature</th>
<th>SRS n=143</th>
<th>SRS n=50</th>
<th>mUPD(7) n=24</th>
<th>r(15) n=2</th>
<th>t(17q25) n=2</th>
<th>Deletion CSH1 n=1</th>
<th>18+ mosaic n=1</th>
<th>Deletion (18p11) n=1</th>
<th>Deletion (8q11-13) n=1</th>
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<tbody>
<tr>
<td>Major</td>
<td></td>
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<td></td>
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<tr>
<td>Low birth weight (&lt;−1 SD)</td>
<td>94%</td>
<td>94%</td>
<td>87.5%</td>
<td>2/2</td>
<td>2/2</td>
<td>1/1</td>
<td>1/1</td>
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</tr>
<tr>
<td>Short stature (&lt;−1 SD)</td>
<td>98%</td>
<td>86%</td>
<td>100%</td>
<td>2/2</td>
<td>2/2</td>
<td>1/1</td>
<td>1/1</td>
<td>0/1</td>
<td>1/1</td>
</tr>
<tr>
<td>Triangular face</td>
<td>79%</td>
<td>&gt;62%</td>
<td>29%</td>
<td>2/2</td>
<td>2/2</td>
<td>1/1</td>
<td>1/1</td>
<td>1/1</td>
<td>Atypical</td>
</tr>
<tr>
<td>Minor</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Clinodactyly V</td>
<td>68%</td>
<td>56%</td>
<td>37.5%</td>
<td>2/2</td>
<td>2/2</td>
<td>0/1</td>
<td>0/1</td>
<td>1/1</td>
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<tr>
<td>Relative macrocephaly</td>
<td>64%</td>
<td>70%</td>
<td>71%</td>
<td>0/2</td>
<td>2/2</td>
<td>1/1</td>
<td>1/1</td>
<td>0/1</td>
<td>0/1</td>
</tr>
<tr>
<td>Ear anomalies</td>
<td>53%</td>
<td>70%</td>
<td>64%</td>
<td>0/2</td>
<td>2/2</td>
<td>0/1</td>
<td>1/1</td>
<td>1/1</td>
<td>Atypical</td>
</tr>
<tr>
<td>Skeletal asymmetry</td>
<td>51%</td>
<td>34%</td>
<td>45%</td>
<td>0/2</td>
<td>2/2</td>
<td>1/1</td>
<td>1/1</td>
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<tr>
<td>Brachydactyly V</td>
<td>48%</td>
<td>25%</td>
<td>25%</td>
<td>0/2</td>
<td>2/2</td>
<td>1/1</td>
<td>1/1</td>
<td>0/1</td>
<td>0/1</td>
</tr>
<tr>
<td>Downward slanting corners of mouth</td>
<td>46%</td>
<td>0%</td>
<td>0%</td>
<td>2/2</td>
<td>1/2</td>
<td>0/1</td>
<td>0/1</td>
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<td>0/1</td>
</tr>
<tr>
<td>Muscular hypotrophy/tonia</td>
<td>45%</td>
<td>12.5%</td>
<td>0.2</td>
<td>0/2</td>
<td>0/2</td>
<td>0/1</td>
<td>1/1</td>
<td>1/1</td>
<td>1/1</td>
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<tr>
<td>Motor/neuropsychological delay</td>
<td>37%</td>
<td>38%</td>
<td>23%</td>
<td>2/2</td>
<td>0/2</td>
<td>1/1</td>
<td>1/1</td>
<td>1/1</td>
<td>1/1</td>
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<tr>
<td>Irregular spacing of teeth</td>
<td>28%</td>
<td>4%</td>
<td>0/2</td>
<td>0/2</td>
<td>0/2</td>
<td>0/1</td>
<td>1/1</td>
<td>1/1</td>
<td>1/1</td>
</tr>
<tr>
<td>Sunan crease</td>
<td>25%</td>
<td>4%</td>
<td>0/2</td>
<td>0/2</td>
<td>0/2</td>
<td>0/1</td>
<td>1/1</td>
<td>1/1</td>
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<tr>
<td>Squeaky voice</td>
<td>22%</td>
<td>4%</td>
<td>0/2</td>
<td>0/2</td>
<td>0/2</td>
<td>0/1</td>
<td>1/1</td>
<td>1/1</td>
<td>1/1</td>
</tr>
<tr>
<td>Syndactyly</td>
<td>19%</td>
<td>4%</td>
<td>0%</td>
<td>2/2</td>
<td>1/2</td>
<td>0/1</td>
<td>1/1</td>
<td>1/1</td>
<td>1/1</td>
</tr>
<tr>
<td>Cafe au lait naevi</td>
<td>19%</td>
<td>4%</td>
<td>4%</td>
<td>1/2</td>
<td>0/2</td>
<td>0/1</td>
<td>1/1</td>
<td>1/1</td>
<td>1/1</td>
</tr>
<tr>
<td>Early or precocious puberty</td>
<td>13%</td>
<td>0%</td>
<td>0%</td>
<td>0/2</td>
<td>1/2</td>
<td>0/1</td>
<td>1/1</td>
<td>0/1</td>
<td>0/1</td>
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<tr>
<td>Genital abnormalities</td>
<td>36%</td>
<td>0%</td>
<td>0%</td>
<td>0/2</td>
<td>1/2</td>
<td>0/1</td>
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<td>Speech delay</td>
<td>20%</td>
<td>25%</td>
<td>0%</td>
<td>0/2</td>
<td>0/2</td>
<td>0/1</td>
<td>1/1</td>
<td>0/1</td>
<td>0/1</td>
</tr>
<tr>
<td>Camptodactyly</td>
<td>22%</td>
<td>4%</td>
<td>0%</td>
<td>0/2</td>
<td>1/2</td>
<td>0/1</td>
<td>1/1</td>
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<td>1/1</td>
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<tr>
<td>Feeding difficulties</td>
<td>56%</td>
<td>33%</td>
<td>0%</td>
<td>0/2</td>
<td>1/2</td>
<td>0/1</td>
<td>1/1</td>
<td>1/1</td>
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References 3 4 5 1, 52, 59 19, 20 28, 29 35 13 17 18
Silver-Russell syndrome (SRS) is a clinically and genetically heterogeneous disorder of growth with a spectrum of additional dysmorphic features. SRS may comprise different disorders with clinically similar phenotypes or may result from disruption of different components of a single biochemical or endocrinological pathway, in either case reflecting its genetic heterogeneity. This review focuses on the influence of genomic imprinting in SRS and recent progress in defining two candidate SRS regions on both the p and q arms of chromosome 7. The investigation of imprinted genes on chromosome 7 and candidate genes from other chromosomal locations are also discussed with regard to their potential role in SRS.

Clinical features of Silver-Russell syndrome
SRS is a congenital disorder characterised by intrauterine and postnatal growth retardation in association with a number of dysmorphic features first described in 1953 by Silver et al and in 1954 by Russell. The syndrome is clinically heterogeneous and diagnosis may be subjective. Two recent reviews provide details of the spectrum and frequency of the various clinical manifestations of SRS in two large groups of patients. Wollmann et al additionally reviewed the growth characteristics of 386 SRS patients. Table 1 lists the clinical features of SRS and their frequencies, as determined in these two reviews. Low birth weight, short stature resulting from postnatal growth retardation, and a characteristic, small, triangular face were observed in the vast majority of patients. Clinodactyly of the fifth finger, relative macrocephaly owing to sparing of cranial growth, and facial, limb, or body asymmetry were also frequently observed. Although no strict diagnostic criteria for SRS have been established, presence of the three major features plus one or more of the minor confirmatory features (such as clinodactyly or asymmetry) are generally required for a positive diagnosis. However, none of these traits is absolute and there are a small number of diagnosed SRS patients in whom a major feature is absent, but several of the minor ones are present. A recent novel observation is the presence of bilateral campodactyly with terminal interphalangeal contractures in 10 of 31 classical SRS patients.

Genetic heterogeneity
The majority of SRS cases are sporadic. However, in a review of 197 SRS probands, 19% had more than one affected person in the family, providing evidence for a genetic cause. Different modes of inheritance have also been suggested on the basis of family histories, indicating that SRS is genetically heterogeneous (reviewed by Duncan et al). Autosomal recessive transmission appears likely in several families in which there is more than one affected sib but the parents are phenotypically normal. Consanguinity between the normal parents in four unrelated Arab families with two or more SRS offspring supports transmission of an autosomal recessive mutation in these cases. Autosomal and X linked dominant inheritance patterns have also been suggested in SRS.

In their review, Duncan et al describe 23 families with 38 SRS patients. In 17 of these, multiple maternal relatives either had full SRS or a partial phenotype. Autosomal dominant inheritance, with intrafamilial variation in expression owing to incomplete penetrance or gene pleiotropy is one possibility. Alternatively, the primarily female transmission of this disorder in these families is suggestive of an X linked dominant form. While paternal inheritance of SRS has been described, no male to male transmission has been documented. Hypogonadism accompanying SRS in males may be a factor in reducing male transmission of SRS. Nevertheless, this means that X linked dominant and autosomal dominant inheritance patterns may be indistinguishable in these families. In view of recent evidence to suggest that genomic imprinting plays a role in certain SRS patients, an imprinted mode of inheritance may be invoked in some SRS families with more than one affected. Certainly the predominant maternal transmission of SRS is compatible with a matrilinear mode of inheritance owing to genomic imprinting.

Concordance for SRS in a pair of monozygotic twins discordant for SRS is confounding. This may be solely because of environmental factors. Placental pathology, including hypoplasia, a single umbilical artery, and velamentous cord insertion was observed in the growth restricted twin fetus during pregnancy. However, the continuation of growth retardation postnatally is difficult to explain if purely environmental circumstances are involved. Alternatively, genetic factors such as a postzygotic mutation event or mosaicism, in concert with environmental factors, may be the cause of the disease phenotype in only one twin.

While genetic factors are evident in familial cases of SRS, the genetic contribution to the majority of isolated patients is difficult to assess. It is possible that some of these patients who are karyotypically normal have inherited recessive mutations, or have new dominant mutations, although there is no evidence of a parental age effect to support the latter.

Structural chromosome abnormalities in patients with SRS or SRS-like features
The majority of SRS patients have a normal karyotype. However, a small number of patients with SRS, or features reminiscent of SRS, have been described with structural abnormalities of different chromosomes, further illustrating the genetic heterogeneity of this disorder. These chromosomal disruptions provide clues regarding the location of the genes involved in SRS, aiding a positional cloning or positional candidate approach to identifying these. Several candidate genes mapping in these regions have been identified and analysed for involvement in SRS.
The size of the deleted sections of distal 15q in r(15) syndrome patients are variable, but where these included IGFIR, the patients were severely pre- and postnatally growth retarded.\textsuperscript{21}\textsuperscript{22}\textsuperscript{24} The insulin-like growth factor (IGF) family, consisting of insulin, IGF1 and IGF2, their receptors IGF1R and IGF2R, and six IGF binding proteins (IGFBP), plays a crucial role in regulation of fetal and neonatal growth and development.\textsuperscript{25}\textsuperscript{26} It was hypothesised that hemizygous loss of the IGFIR gene was responsible for the phenotype in the two SRS patients with r(15) and the shared features between SRS and r(15) syndrome patients. IGFIR was also proposed as a candidate for SRS in patients with normal karyotypes.\textsuperscript{20} However, no hemizygous deletions were identified in a total of 38 such SRS patients, screened in two separate studies.\textsuperscript{24}\textsuperscript{27} Furthermore, no mutations were identified in two exons encoding crucial active sites within the protein in 33 probands, indicating that this gene cannot be largely responsible for SRS in other groups of patients.\textsuperscript{27}

**DISTAL CHROMOSOME 17q**

Two patients with severe SRS have been described with reciprocal translocations involving distal chromosome 17q, with the breakpoints originally localised to 17q25.\textsuperscript{28}\textsuperscript{29} Both patients had several minor SRS traits in addition to the three major criteria (table 1). The first case had an apparently balanced translocation (17;20)(q25;q13) inherited from the phenotypically normal father.\textsuperscript{20} The second case had a de novo translocation (1;17)(q31;q25).\textsuperscript{29} The translocation breakpoint in the latter case has now been cloned and the localisation refined to 17q23.3-q24.\textsuperscript{30}

Two members of the growth factor receptor bound protein (GRB) family of genes, GRB2 and GRB7, which map to 17q21.1 and 17q21.1 respectively, have been analysed as candidates for SRS. The GRB proteins interact with various receptor tyrosine kinases within the insulin and IGF1 signal transduction axes, and so play a role in mitogenesis (reviewed by Daly et al\textsuperscript{31}). GRB2 and the guanine nucleotide exchange factor, Sos, form a complex, which interacts with the insulin receptor substrate I (IRS-I), and thus regulates Ras activation in the first steps of the insulin signalling pathway.\textsuperscript{32} Minor structural anomalies of GRB2 and GRB7 have been excluded in 36 SRS patients with normal karyotypes (M Hitchins, unpublished data). GRB2 was screened for mutations of the coding region by two groups, in a total of 29 SRS patients, but none was found, indicating that these genes do not play a significant role in SRS (M Hitchins, unpublished data). However, the potential effects of the two reported translocations on expression of these genes has not been investigated. The growth hormone (GH) gene cluster, including the GH gene and the chorionic somatomammotropin hormone (CSH) genes CSH1 and CSH2, are located on chromosome 17q24.1.\textsuperscript{34} A patient with classical SRS (table 1) has been reported with a paternally inherited deletion of the CSH1 gene.\textsuperscript{35} Recently, a second case with a
similar deletion has been identified (T Eggermann, personal communication). CSH1, or placental lactogen, is produced in the placental syncytiotrophoblast and secreted into the fetal and maternal circulation. Low levels of placental lactogen during pregnancy have been associated with pathological conditions including IUGR, diabetes, and pre-eclampsia, but these may be because of associated factors such as placental infarction restricting maternal-fetal circulation (reviewed by Handwerger\textsuperscript{36}). Several cases with normal phenotypes at birth have been reported with absence of placental lactogen during pregnancy owing to CSH1 deletions on both chromosome 17 homologues in the fetus.\textsuperscript{37} 38 Thus, the significance of the hemizygous CSH1 deletion in the two SRS patients is unclear, but may have contributed to the syndrome in combination with other genetic or environmental factors. Infarction of the placenta, for instance, was noted in the first case.\textsuperscript{39} Deletion of CSH1 in association with SRS is rare and similar deletions have been ruled out in 106 SRS patients to date (T Eggermann, personal communication; M Hitchins, unpublished data).

Chromosome 7 and genomic imprinting in SRS

UNIPARENTAL DISOMY AND GENOMIC IMPRINTING

Uniparental disomy (UPD) is the rare inheritance of both copies of one particular chromosome from a single parent.\textsuperscript{40} UPD occurs through two main mechanisms, following a non-disjunction event during meiosis. (1) Fusion of a disomic gamete with a normal monosomic one, with subsequent loss of one of the extra homologues from the trisomic conceptus, is termed “trisomic rescue”. Uniparental heterodisomy, which is the inheritance of two different homologues from one parent, that is, both grandparental copies, arises through this mechanism. This may also be accompanied by the observation of trisomy mosaicism in the placenta.\textsuperscript{41} (2) Fusion of a nullisomic gamete with a normal one forming a monosomic zygote, followed by mitotic duplication of the existing homologue in the conceptus, is termed “monosomy rescue”. This mechanism results in two replica copies of the original parental homologue, referred to as uniparental disodismy. UPD may be a mixture of hetero- and isodisomy owing to recombination at meiosis I. UPD can also occur through a mitotic error after fertilisation. This may result in segmental disomy following somatic recombination, or may be accompanied by trisomy mosaicism or partial supernumerary chromosomes.\textsuperscript{42-43}

Genetic disorders as a consequence of UPD may occur through two distinct mechanisms. (1) Recessive mutations can be reduced to homozygosity owing to isodisomy of the locus if the transmitting parent is a heterozygous carrier. (2) Disruption of expression of genes subject to genomic imprinting can occur, in both iso- and heterodisomy, where the chromosome in question contains an imprinted region.

Genomic imprinting is defined as the differential expression of a gene or chromosomal region according to the parental origin of inheritance. Imprinted genes are expressed from a single parental allele (fig 1A), such that in UPD there may be absence of an active copy on the parental homologue that is missing, or overexpression on the duplicate parental chromosome (fig 1B). Imprinted genes tend to be clustered together in regions in which their expression is co-ordinately regulated and are associated with sites of parental allele specific methylation of the DNA. A regularly updated catalogue of imprinted genes may be found at www.otago.ac.nz/IGC.\textsuperscript{44} Imprinted genes and regions are also generally conserved in evolution. Imprinted regions in the mouse have been defined by the use of reciprocal translocations to derive embryos that are uniparental for specific subchromosomal regions, which give rise to a noticeable phenotype.\textsuperscript{45} There are currently 14 defined regions of “non-complementation”, which require both parental complements for normal growth and development, spread over eight different mouse chromosomes. A map of these imprinted regions and genes in the mouse, with the human homology map, is available at www.mgu.har.mrc.ac.uk/imprinting/imprinting.html.

MATERNAL UNIPARENTAL DISOMY FOR CHROMOSOME 7 AND SRS

Maternal UPD for chromosome 7 (mUPD(7)), that is, the inheritance of both homologues from the mother, occurs in 7-10% of SRS cases.\textsuperscript{46 47} The first cases of mUPD(7) were ascertained through diagnosis of recessive disorders, but prompted researchers to screen for UPD(7) among the SRS population. Two cases with cystic fibrosis (CF) resulting from maternal isodisomy for chromosome 7, for which the mothers were CFTR mutation carriers, were the first cases of UPD to be described.\textsuperscript{48 49} Both patients had postnatal growth retardation, which could not be attributed to CF alone, and other features of SRS. A third patient with a collagen disorder and
marked short stature was found to have mixed
maternal hetero- and isodisomy for chromosome 7, with a homozgyous mutation of the
COLIA2 gene.40 Screens of entire SRS patient
cohorts for mUPD(7) have subsequently been
carried out and at least 40 cases of mUPD(7)
in association with SRS (or SRS features) have
been published.40 43 46–56 Some researchers ex-
inclusion mUPD(7) in their group of patients,57
which helped in determining the frequency of
this occurrence more accurately. UPD involv-
ing chromosomes 2, 6, 9, 14, 16, 20, and 22 in
association with SRS have also been investi-
gated, but ruled out.58

Some cases of SRS with mUPD(7) have a mild or incomplete phenotype.47 59 Features
appearing milder or less frequently in
mUPD(7) patients include the facial dysmor-
phism, asymmetry, clindactyly and other di-
gital abnormalities, cafe au lait spots, precocious puberty, squeaky voice, and tooth anomalies, as
listed in table 1.31 52 59 However, mUPD(7)
patients experienced severe feeding difficulties in the first few years of life, speech delay, and
excessive sweating. These patients may com-
prise a more homogeneous group that are
clinically distinguishable from other SRS pa-
tients.4 47 59

MECHANISM CAUSING SRS IN mUPD(7)
The mechanism by which mUPD(7) causes
SRS has been investigated. Cases with biparen-
tinal inheritance of chromosome 7, but with con-
fined placental mosaicism for trisomy 7 as a
consequence of “trisomic rescue” were found
to have normal birth weights. This excluded the
possibility that IUGR in SRS is the result of
malfucntion of a trisomy 7 placenta.60 Two patients with paternal UPD(7) (pUPD(7))
have been reported with recessive disorders
owing to isodisomy at mutant recessive loci.
The first patient had congenital chloride
diaryseh but had normal growth patterns.60
The second child had CF with complete situs
inversus and immotile cilia in addition, and had
door growth at 6 months of age most likely as a
consequence of his recessive phenotype.61 The
lack of a distinct growth phenotype in the
patients with pUPD(7), in contrast to the con-
sistent severe growth retardation seen in
mUPD(7), implicates imprinted genes in the
diagnosis of SRS in these cases.

To differentiate between unmasking of a
recessive allele by isodisomy and disruption of
genomic imprinting as the cause of SRS, Preec et al41
analysed the full length of the
chromosome 7 homologues in five mUPD(7)
cases for regions of consistent isodisomy.11
Forty polymorphic markers distributed along
the entire chromosome 7, at an average genetic
interval of 4.5 cM, were analysed in each
patient. The chromosmes 7 were mixed hetero- and isodisomy in each case, but no
region of isodisomy was common to all five
patients. This study excluded recessive muta-
tions as the cause of SRS in cases with
mUPD(7), indicating that an imprinting defect
is the most likely cause for this disorder.11 SRS
could be caused by the absence of expression of
a gene involved in growth promotion that is
active exclusively on the paternal allele, or by
excess of a maternally expressed imprinted
gene that is involved in growth inhibition (fig
1B).

Two separate candidate regions for SRS
on chromosome 7
Maternal UPD(7) in SRS has involved the
whole chromosome in the majority of cases.
Thus, until recently, regions of human chro-
mosome 7 that were homologous to imprinted
regions in the mouse provided the main clue as
to the location of imprinted genes that were
likely implicated in this disorder. Different segments of human chromosome 7 share homology with regions
from 10 different mouse chromosomes includ-
ing 2, 4, 5, 6, 7, 10, 11, 12, 13, and 16. These
are detailed at www.ncbi.nlm.nih.gov/
Homology/human7.html. However, just two
regions on human 7 share homology with definitive mouse imprinted regions; human
7p11.2-p13 is homologous to mouse proximal
chromosome 11 and human 7q21-qter has a
conserved region on proximal mouse chromo-
some 6 (fig 2). Both these regions in mice are
associated with growth phenotypes. These
regions aid in the selection of candidate genes
for SRS and enable the study of imprinting
processes that may contribute to SRS in the
mouse system.

Recently the identification of key SRS
cases with anomalies of 7p11.2-p13 and
7q31-qter has defined these two intervals as
separate candidate regions for SRS. It is inter-
esting to note that these two segments share
homology with the aforementioned mouse
imprinted regions.

THE 7p11.2-p13 REGION AND CANDIDATE GENES
Two patients with SRS have been identified
with duplications of 7p11.2-p13.62 63 In the first
case, the proband and her mother had an
inverted interstitial duplication of this region.
The SRS proband had relatively low birth
weight, marked short stature, mild facial asym-
smetry, and fifth finger clinodactyly. The
mother, whose duplication had arisen de novo
on the grandpaternal chromosome, also had
mild features of SRS, including short stature
and fifth finger clinodactyly.63 The second
report was of a de novo tandem duplication of
the region 7p11.2-p13 on the maternally
derived homologue, in a SRS patient present-
ing with pre- and postnatal growth restriction,
small triangular face, and fifth finger clinodac-
tyly, but no asymmetry.63 In both cases the
duplicated region encompassed the insulin-like
growth factor binding protein 1 (IGFBP1) and
the growth factor receptor bound protein 10
gene (GRB10). The epidermal growth factor
receptor gene (EGFR) lay outside the duplica-
tions (fig 2).62 63 Each of these genes had previ-
ously been proposed as candidates for SRS as
they are important in the regulation of growth
and development and their murine homo-
logues may within the conserved imprinted
region on proximal mouse chromosome 11.

Mice with maternal disomy/duplication for
proximal 11 (MatDp.prox11) are 30% smaller
than their normal littermates, possibly representing the growth restriction seen in SRS. Conversely, mice with paternal disomy for this region (PatDp.prox11) are 30% larger than normal. Although pUPD(7) is not associated with a growth phenotype in humans, there are other imprinted genes within proximal mouse 11 whose human homologues map to chromosomes other than 7, which may account for the overgrowth in PatDp.prox11 mice. For example, the paternally expressed U2af1-rs1 gene with a human equivalent U2AF1-RS1 on chromosome 5 is a candidate for this excessive growth in these mice (fig 2).

EGFR was previously shown to be an unlikely candidate for SRS on the basis that it was not imprinted during human fetal development. That EGFR is not within the duplicated region in the two recently described SRS patients corroborates this (fig 2). Other imprinted genes within proximal mouse 11 whose human homologues map to chromosomes other than 7, which may account for the overgrowth in PatDp.prox11 mice. For example, the paternally expressed U2af1-rs1 gene with a human equivalent U2AF1-RS1 on chromosome 5 is a candidate for this excessive growth in these mice (fig 2).

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in fetal skeletal muscle, which was maternally expressed. In contrast, \textit{GRB10} was paternally expressed in fetal brain and spinal cord, but biallelic in all other major fetal organs and tissues. The role of \textit{GRB10} may play in SRS is difficult to discern on the basis of this imprinting profile. Yoshihashi et al. reported a maternally transmitted P9SS amino acid substitution in \textit{GRB10} in two unrelated Japanese SRS patients, but this was not found in a further 161 SRS subjects. This substitution was subsequently shown to be a rare polymorphism in the Japanese population and unlikely to be causative of SRS. No mutations of \textit{GRB10} have been identified in a total of 139 SRS patients screened for the entire coding region of the gene, indicating that it does not contribute significantly to SRS. Disruption of imprinted expression of \textit{GRB10}, through altered methylation for instance, has not been investigated and remains a possibility. Since imprinted genes tend to occur in clusters, it is likely that other imprinted genes exist in the 7p11.2-p13 candidate region, which may be responsible for SRS in cases with and without mUPD(7).

Different opinions regarding whether the SRS phenotype in patients with a duplication of 7p11.2-p13 is attributable to an imprinting effect have been put forward. Monk et al. stated that duplication of this region in SRS provided evidence for involvement of a maternally expressed growth suppressor gene, as opposed to a growth promoter gene active on the paternal allele, assuming these cases have a common aetiology with mUPD(7) patients. However, others have suggested that SRS may be instead the result of an extra copy of this region. The 7p12.1-p13 duplication in the SRS proband described by Joyce et al. was of paternal origin in the proband’s mother, who also had SRS characteristics. However, the mother’s clinical phenotype was mild and may have been caused by duplication or disruption of non-imprinted contiguous genes, which may contribute to some of the minor features associated with SRS. A SRS patient with maternal isodisomy 7 and ring chromosome 7 (r(7)) has been reported. The r(7) consisted of the 7p11-q11 region and was identified as paternal in origin. It is most likely that mitotic duplication of the maternal 7 homologue occurred in the zygote that was essentially monosomic, with just a partial paternal chromosome 7 to correct for chromosome number. Mosaicism for the r(7) probably reflected the instability of supernumerary partial chromosomes during cell replication. Although mUPD(7) was considered the underlying cause of SRS, the authors suggested that genes in the 7p11-q11 region were not involved in SRS since a paternal copy was present. However, the behaviour of genes in ring chromosomes is not clear, and so a paternal copy in some cells may not compensate fully for loss of a complete paternal homologue. It is plausible that partial trisomy for chromosome 7 may be responsible for aspects of the SRS phenotype in this patient, in the same way that trisomy mosaicism for distal chromosome 11 may cause hemihypertrophy in Beckwith-Wiedemann syndrome patients with segmental pUPD for 11p15.5. No evidence for low level trisomy 7 mosaicism has been identified in four SRS patients with mUPD(7), though further mUPD(7) patients should be screened, especially those with asymmetry, before this can be ruled out as a contributory mechanism for SRS.

THE 7q31-qter REGION AND CANDIDATE GENES

Two patients with SRS or some of the major SRS features have been reported with mUPD restricted to the q arm of chromosome 7. This suggests that the imprinted genes responsible for SRS in patients with mUPD(7) for the entire chromosome are located on the q arm and that the p arm may not involve an imprinting aetiology. A unique patient was reported with isochromosomes of 7p and 7q, showing homozygosity at all chromosome loci investigated. Paternal isodisomy was found for the 7p arm and the 7q arm was maternally isodisomic. The patient did not have IUGR, but became growth retarded postnatally, had mild limb asymmetry, slight fifth finger clinodactyly, and a triangular shaped face. The authors hypothesised that a maternal contribution to the p arm was necessary for normal fetal growth in utero, but that imprinted genes on the q arm were responsible for the additional SRS features noted in this patient. Recently, a patient with typical SRS including severe IUGR, short stature, relative macrocephaly, slightly triangular face with downturned corners of the mouth, mild fifth finger clinodactyly, abnormal spacing of the teeth and a squeaky voice, but no asymmetry, was described with segmental maternal isodisomy for the region 7q31-qter. This report shows that mUPD for this small interval on chromosome 7 causes a clinically consistent SRS phenotype, including several of the minor features. The finding of mUPD restricted to 7q31-qter in this SRS patient indicates that two separate regions on chromosome 7 can independently cause SRS, with the telomeric region of the q arm most certainly involving an imprinting defect (fig 2).

The human 7q21-qter region is homologous to proximal mouse chromosome 6, which shows two distinct imprinting phenotypes in mice with maternal UPD. These are defined by two translocation breakpoints, with the proximal region between the centromere and the T77H breakpoint, and the distal region between the T77H and T6Ad translocation breakpoints (fig 2). The distal imprinting segment is associated with pre- and postnatal growth restriction in mice with maternal disomy. This region contains a cluster of imprinted genes including the paternally expressed mesoderm specific transcript gene (\textit{Mest}/\textit{Peg1}) and the nonclathrin-coatprotein gene (\textit{γ-2Cop}), which is maternally expressed in mouse, in contrast to its human homologue. Two untranslated paternally expressed transcripts \textit{Mit1} and \textit{γ-2Cop} antisense, which overlap \textit{γ-2Cop}, have also been reported in mouse. \textit{Mest} was first identified in mice by subtractive
hybridisation of cDNAs from a normal mouse embryo with those from a parthenogenetic embryo (with just a maternal genetic component) in a screen for paternally expressed genes. Mice with a paternally inherited mutation of the Mest gene have IUGR and the female offspring themselves show abnormal maternal behaviour, with impaired placentophagia. The human homologue MEST has thus been proposed and investigated as a candidate for SRS.

Human MEST and \( \gamma \)-2 COP both map to 7q32 (fig 2). MEST is paternally expressed in embryos and all major fetal organs, but was found to be biallelic in adult lymphocytes. Two different isoforms of MEST have since been identified in lymphocytes, one of which is biallelically expressed while the other is transcribed specifically from the paternal allele. Functional absence of MEST was shown in SRS patients with mUPD(7) by analysing this latter isoform in patient lymphoblasts. However, no mutations of MEST have been identified in 49 SRS patients. MEST has a differentially methylated 5' CpG island, thought to regulate expression, with hypermethylation of the inactive maternal allele. Although patients with mUPD(7) only have the methylated maternal alleles at this site, no epigenetic alterations were identified in 35 non-mUPD(7) SRS patients, suggesting that their regulation of MEST was normal. These data indicated that MEST is unlikely to play a major role in SRS, although it may still contribute to the phenotype in mUPD(7) patients. The \( \gamma \)-2 COP gene is paternally expressed in all major fetal organs, with the exception of brain and liver, where biallelic expression was noted. \( \gamma \)-2 COP was screened for mutations in 42 SRS patients. A substitution was observed in one of these patients, which was not identified in 98 normal subjects, but this was present on the inactive maternal allele. A non-translated paternally expressed transcript, CIT1, contained within an intron of \( \gamma \)-2 COP, has also recently been reported. Intensive physical and transcript mapping within 7q32 will help to identify other genes which may be subject to imprinting in this region.

Is 7q21 a novel imprinted region?

A novel imprinted region may exist at 7q21. This is suggested by the homology between this region and the proximal imprinted region on mouse chromosome 6, between the centromere and T77H translocation breakpoint (fig 2). Maternal disomy for this region causes early embryonic lethality. This segment contains the paternally expressed \( \gamma \)-sarcoglycan gene (SGce). The human homologue SGCE maps to 7q21, but its imprinting status is unknown. Furthermore, a paternally expressed gene, PEG10, has recently been identified which maps to 7q21 near to SGCE. PEG10 appears to be derived from a retrotransposon. Although these genes may be located within a third imprinted region on chromosome 7, this lies proximal to the candidate imprinted region defined by segmental mUPD of 7q31-qter in SRS, and no chromosomal anomalies involving 7q21 have been associated with SRS to date.

Conclusions

The Silver-Russell syndrome is both clinically and genetically heterogeneous, with different modes of inheritance and chromosomal locations involved. This makes identification of the genes responsible difficult. Recent progress has been made on chromosome 7, defining two separate candidate regions, 7p11.2-p13 and 7q31-qter. A definitive role for genomic imprinting has been shown in the 7q31-qter region, as a result of the observation of maternal uniparental disomy specifically for this interval in a single SRS patient. This case suggests that the 7q31-qter region alone may be responsible for SRS in patients with mUPD for the entire chromosome 7. Two imprinted genes within the disomic region, MEST and \( \gamma \)-2 COP, have been investigated as candidates for SRS, but there has been no direct evidence to show either is responsible for the phenotype. Currently there are conflicting opinions regarding the underlying basis for SRS in patients with structural anomalies of the 7p11.2-p13 region, with some arguing against a role for imprinting. However, this region contains one imprinted gene, which has been implicated in SRS, OR18, though its potential involvement in the disorder remains uncertain. It may be the case that both the 7q31-qter and 7p11.2-p13 candidate regions cause SRS through disruption of imprinting. Since imprinted genes tend to be clustered in groups, there are likely to be other imprinted genes in both chromosome 7 candidate regions, which will require investigation for a role in SRS. The identification and analysis of additional SRS patients presenting with unique chromosomal abnormalities involving chromosome 7 will further define the candidate intervals. It is plausible that, as a variable phenotype, no single gene is responsible for all the features of SRS in any person. SRS may be a contiguous gene disorder, or where imprinting is involved, be the result of disruption of expression of several co-regulated imprinted genes within an imprinted region. Identification of the gene or genes causing SRS in one of the associated chromosomal locations (in particular on 7p, 7q, or chromosome 17) should aid considerably in the identification of the other genes involved on different chromosomes, by providing information on the growth and developmental pathway affected.

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