Failure of homologous synapsis and sex-specific reproduction problems

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INTRODUCTION

Down syndrome is the most common chromosomal abnormality that manifests in newborns. The karyotype of 90% of Down syndrome cases is standard trisomy 21, whereas in approximately 4% of cases an extra chromosome 21 is associated with the Robertsonian translocation of chromosome 21 (Gardner and Sutherland, 2004). About three-quarters of Down syndrome cases with Robertsonian translocation occur de novo, while the remaining one-quarter are familial cases in which either parent is a balanced carrier of the same translocation. In these familial cases, a distinct bias in the transmission of the translocation occurs: the balanced carrier of the translocation is almost always the mother (Gardner and Sutherland, 2004). Likewise, reciprocal translocation between autosomes shows similar transmission bias. Constitutional t(11;22)(q23;q11.2) translocation is a well-known recurrent reciprocal translocation. The only viable unbalanced carrier of the same translocation. In these familial cases, a distinct bias in the transmission of the translocation occurs: the balanced carrier of the translocation is almost always the mother (Gardner and Sutherland, 2004). Likewise, reciprocal translocation between autosomes shows similar transmission bias. Constitutional t(11;22)(q23;q11.2) translocation is a well-known recurrent reciprocal translocation. The only viable unbalanced carrier of the translocation is the male.

The prophase of meiosis I ensures the correct segregation of chromosomes to each daughter cell. This includes the pairing, synapsis, and recombination of homologous chromosomes. A subset of chromosomal abnormalities, including translocation and inversion, disturbs these processes, resulting in the failure to complete synapsis. This activates the meiotic pachytene checkpoint, and the gametes are fated to undergo cell cycle arrest and subsequent apoptosis. Spermatogenic cells appear to be more vulnerable to the pachytene checkpoint, and male carriers of chromosomal abnormalities are more susceptible to infertility. In contrast, oocytes tend to bypass the checkpoint and instead generate other problems, such as chromosomal imbalance that often leads to recurrent pregnancy loss in female carriers. Recent advances in genetic manipulation technologies have increased our knowledge about the pachytene checkpoint and surveillance systems that detect chromosomal synapsis. This review focuses on the consequences of synapsis failure in humans and provides an overview of the involved pathways that leads to the differences in reproductive outcomes between males and females.

Keywords: synapsis, pachytene checkpoint, translocation, sex body, meiotic sex chromosome silencing, meiotic silencing of unsynapsed chromosomes, Homed1

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first is that a part of the male translocation carrier population does not transmit the translocation to his offspring because these car-
riers are infertile due to azoosperma. Females and men without azoosperma might suffer from RPL but can also produce off-
spring with either of normal chromosome, balanced translocation or unbalanced. Indeed, a family with autosomal translocation and
male-only sterility has been reported, in which the female carrier had two consecutive spontaneous abortions (Paoloni-Giacobino et al., 2000). This might exert a large impact on the female bias of transmission. Another factor that affects bias is transmission
distortion of the translocation chromosomes. In general, female translocation carriers are more frequently transmit the translo-
cation to the offspring than male carriers without azoosperma (Gardiner and Sutherland, 2004). This latter phenomenon might be independent of the former, but the nature of this phenomenon is less understood.

In this review, we focus on chromosomal translocation and male-only sterility. Recently, mutations in the SYCP3 gene were
reported in RPL women experiencing more than three sponta-
neous abortions during early gestation (Bolot et al., 2009). The
mutant genes consistently encode C-terminally mutated proteins
that might inhibit the normal function of the SYCP3 protein via
dominant negative effects and have been predicted to generate
aneuploid oocytes. Interestingly, a mutation in a similar region
in the SYCP3 gene was also reported in azoosperma patients
with meiotic arrest (Miayamoto et al., 2003). These data indi-
cate that the quality controls of meiotic chromosome segregation
are more stringent in males than in females (Hunt and Hassold, 2002). "Checkpoint" is defined as a cellular surveillance system
that detects abnormalities and is dispensable when everything is
normal. It has been speculated that the sexual dimorphism of these
reproductive outcomes might be due to differences in checkpoint
robustness between males and females (Kurahashi et al., 2009).

MEIOTIC ARREST OF SPERMATOGENESIS IN THE MALE
TRANSLOCATION CARRIER

There is an emerging consensus that the events that occur during
the prophase of meiosis I are essential for the proper segrega-
tion of homologous chromosomes (Hassold et al., 2000; Handel
and Schimenti, 2010). Homologous chromosomes that behave
independently during mitotic division have to segregate into
two different daughter cells during meiosis I (Figure 1A). To
accomplish this process, homologous chromosomes interact with
each other, utilizing a specialized pathway known as homol-
ogous recombination (HR; Figure 1B). Initially, programmed
double-strand-breaks (DSBs) manifest in chromosomal DNA by
the action of SPO11 endonuclease at more than 100 sites through-
out the entire genome. Activation of the DSB sites is controlled by
PRDM9, which encodes a H3K4 methylase. PRDM9 binds to the
13 bp recombination hotspot consensus sequence and facilitates
DSB formation via methylation of surrounding histones (Boode
et al., 2009; Baard et al., 2009; Baudat et al., 2010; Myers et al., 2010; Parvanova et al., 2010). To correctly repair these DSBs, a subsequent
HR pathway is activated and the broken DNA ends begin to look
for homologous regions. As the consequence, two homologous
chromosomes are brought together in close association, a process
known as homolog-pairing. A proteinaceous structure known as
the synaptonemal complex (SC) is subsequently formed between
the paired homologous chromosomes. This step is called synapsis.
The DNA lesions are subsequently repaired via HR with the aid of
recombination proteins RAD51 and DMC1. During the final
step of HR, a four-stranded DNA structure, the Holliday junc-
tion that physically connects the two chromosomes is resolved in
one of two ways, crossover or non-crossover. Crossover main-
tains the physical linkage of the chromosomes (chiasmata) and
produces the appropriate bi-orientated tension at the opposite spin-
dle poles during metaphase of meiosis I. Thus, the number and
location of the crossover events is strictly regulated (crossover
assurance and interference). Meiotic recombination, which is well
known as a mechanism that shuffles genetic material in order to
produce variation among individuals, is also indispensable for
the proper segregation of homologous chromosomes (Kurahashi
et al., 2009). Failure of each of these processes triggers cell cycle
arrest and the subsequent apoptosis of meiotic cells (Hochwagen
and Amon, 2006).

The developmental stage during which the SC between the
homologous chromosomes is established is called the pachytene.
In testicular specimens from infertile men with chromosomal
translocations, meiotic arrest and massive cell death at pachytene
have been consistently observed (Solari, 1999; Egozcue et al.,
2006). The spermatocytes appear to be eliminated by a qual-
ity control system, known as the pachytene checkpoint (Roeder
and Ballis, 2000). Obviously, the pachytene configuration of the
homologous chromosomes is considerably affected in the pres-
ence of translocated chromosomes in the translocation carrier.
In the case of reciprocal translocation, homologous pairing can
be theoretically achieved by the formation of a quadrivalent (the
trivalent of the Robertsonian translocation; Figure 1C). However,
the pachytene configuration of translocated chromosomes is vari-
able and partly dependent on the locations of the translocation
breakpoints. The most likely configuration is the quadrivalent,
while asynapsis, partial synapsis, and heterosynapsis between non-
homologous chromosomes have also been observed. Even in the
quadrivalent, full synapsis is not always achieved and small but
substantial portions around breakpoints are often unsynapsed.
The degree of synapsis failure most likely affects the consequences
of the gametes (Martin, 2008; Burgoyne et al., 2009).

DSB-DEPENDENT AND -INDEPENDENT PACHYTENE
CHECKPOINTS

The mechanism of the pachytene checkpoint has been long stud-
ied using yeast, but recent advances in the genetic manipulation
of mice have also revealed the presence of the pachytene checkpoint
in mammals. Over the past decade, a number of knockout mice
that were deficient in genes involved in the meiotic HR pathway
have been created. Because some of the HR proteins are com-
monly utilized in the DSB repair machinery of mitotic cells, the
mutants of these genes manifest as extragonadal phenotypes such
as increased cancer susceptibility (Matzuk and Lamb, 2002; Cohen
et al., 2006). However, regardless of whether the gene is meiosis-
specific or not, infertility is a common phenotype in both male
and female mutants. Defects in HR, e.g., defects in the DNA
repair system, leave unpaired DSBs that most likely trigger the
checkpoint system (Hochwagen and Amon, 2006). Hence, most of
meiotic recombination mutants manifest as meiotic arrest due to the DSB-dependent pachytene checkpoint, leading to azoospermia and oocyte loss by massive apoptosis. In the testicular specimens from Dmc1-deficient mice, extensive apoptosis is consistently observed in stage IV of the seminiferous tubules that mostly includes pachytene spermatocytes (Pittman et al., 1998; Yoshida et al., 1998; de Rooij and de Boer, 2003). In the ovaries of mutants, no follicles have been detected, suggesting that oocytes cannot develop into the subsequent dictyate stage due to extensive apoptosis at the pachytene checkpoint. These data suggest that the DSB-dependent pachytene checkpoint operates similarly both in males and females.

Genetic defects related to SC also manifest as reproductive failure as their only phenotype. These genes do not directly take part in the HR pathway, but defects in SC formation generally impair DSB repair efficiency, which may also activate the DSB-dependent pachytene checkpoint, leading to meiotic arrest (Handel and Schimenti, 2010). However, in such mutants, phenotype severity often differs between males and females but is consistently less severe in females. For instance, knockout mice for genes that encode the components of SC, such as Sycp2 and Sycp3, manifest as male infertility by meiotic arrest, whereas female mice demonstrate subfertility and a variable degree of fetal lethality in utero (Yuan et al., 2000, 2002; Yang et al., 2006). It is still possible that reproductive failure is not simply due to the activation of the DSB-dependent pachytene checkpoint and that some different pathways might be able to cope with SC defects.

![Image](http://example.com/figure1.png)
On the other hand, several lines of evidence suggest that in a subset of mutants, the meiotic phenotype appears to be driven by a DSB-independent pachytene checkpoint. Spo11 initiates meiotic recombination by catalyzing the programmed DSBs. Both male and female Spo11-deficient mice are made sterile by meiotic arrest during the pachytene stage (Baudat et al., 2000; Romanienko and Camerini-Otero, 2000). Mutants demonstrate defects in homologous pairing due to the absence of DSBs and the subsequent development of extensive synopsis failure with some degree of heterosynapsis. Given the lack of DSBs, checkpoints for unrepaired DSBs do not contribute to meiotic arrest. Thus, the pachytene checkpoint in mutants is most likely activated in a DSB-independent manner. Thorough examinations have revealed that putative DSB-independent meiotic arrest is a bit later and less severe than the DSB-dependent (Barchi et al., 2005; Di Giacomo et al., 2005). Similar observations have also been obtained in other DSB-deficient mutant mice (Reinholdt and Schimenti, 2005). These data suggest the possibility of DSB-independent synopsis checkpoints in mammals.

Next, we set out to determine which mechanism is involved in the massive cell death that occurs during pachytene in human males with translocation, the DSB-dependent or -independent pachytene checkpoint. Because synopsis failure can induce meiotic recombination defects, partial synopsis failure due to translocation might instigate enough recombination efficiency to activate the DSB-dependent checkpoint, leading to cell death during the pachytene as well. Indeed, a reduced number of MLH1 foci, a marker of crossover sites at the pachytene, was reported in the spermatocytes of infertile male translocation carrier (Sun et al., 2005; Ferguson et al., 2008; Leng et al., 2009). However, this might not have much of an effect on the fate of the gametes because only the cells harboring DSBs around the breakpoints might have difficulty initiating DSB repair due to functional SC defects. Further, DSB-dependent checkpoints, which appear to operate similarly in males and females, cannot account for the sexual dimorphism of the reproductive outcomes. Thus, the DSB-dependent checkpoint is unlikely to exert a profound effect on the partial synopsis of pachytene chromosomes in translocation carriers.

**ETIOLOGY UNDERLYING SEX DIFFERENCES: INVOLVEMENT OF THE SEX BODY**

How are the chromosomes that undergo partial synopsis subsequently processed? A good example can help imagine this process: the behavior of sex chromosomes during male meiosis. In general, because male sex chromosomes are heteromorphic with small homologous regions known as pseudoautosomal regions at both chromosomal ends, they can make a pair and undergo HR to form the obligatory chiasma required for correct segregation during meiosis I. However, male sex chromosomes cannot be fully synapsed throughout the X or Y chromosome-specific regions and, instead, form a specialized nuclear territory known as the sex body or XY body (Figure 2A, Handel, 2004). The sex body is typically found at the periphery of the nucleus with synapsed chromosomal ends that are anchored to the nuclear lamina. In females, the meiotic stage progress from the pachytene to the subsequent diplotene stage after almost all of the chromosomes are fully synapsed and all of the DSBs are repaired. In contrast, because sex chromosomes cannot be fully synapsed and the DSB repair is delayed in males, the sex body is likely to mask the unsynapsed and unrepaired regions of chromosomes.
of the male sex chromosomes and facilitate cell cycle progression to the subsequent stage despite the presence of unsynapsed and unrepaired chromosomes.

Within the sex body, unsynapsed regions of the male sex chromosomes are transcriptionally inactivated and a number of heterochromatin-associated proteins are assembled into this specialized chromatin domain. This phenomenon is called meiotic sex chromosome inactivation (MSCI, Turner, 2007; Yan and McCarthy, 2009). Similar to X chromosome inactivation in terms of dosage compensation in the female, MSCI is characterized by several epigenetic modifications. For example, the phosphorylated variant of histone H2AX (γH2AX) is detected at sites of MSCI. Other possible suppressive modifications detected include substitution of H3 by variant H3.3 and enrichment of H2A variant, macro-H2A1 (Barends and Grootegoed, 2003; Hoyer-Fender, 2003). MSCI also involves acquisition of other histone modifications, including H2A and H3K9 ubiquitination, with localization of small ubiquitin-like modifier proteins to the sex body (Kata and Feil, 2010). Although this inactivation is transient during prophase I, and the transcription of some of the genes on sex chromosomes restarts after the completion of meiosis, most sex chromosomes genes remain inactivated through late spermatogenesis within a similar nuclear domain called the post-meiotic sex chromatin (Nakagawa et al., 2006; Turner et al., 2006). This process might serve to mark the parental origin of the paternal X chromosome in the zygote. It is still unclear why MSCI occurs. One possible explanation is to prevent the transcription of template chromosomes with unrepaired DSBs, because DSB repair is delayed in unsynapsed sex chromosomes (Cloutier and Turner, 2010; Inagaki et al., 2010). Indeed, mediator of DNA damage checkpoint 1 (MDC1), which binds to the DSB-induced γH2AX, recruits ATR kinase and induces chromosome-wide spreading of the γH2AX leading to the MSCI (Ichijima et al., 2011).

To date, the meiotic behavior of translocated chromosomes has been extensively studied by light microscopic and electron microscopic examinations of the SC in the testicular tissues of infertile men with balanced translocation. A large body of data has been accumulated, demonstrating that translocated chromosomes are often located near the sex chromosomes and occasionally interacted with them (Figure 2B; Luciani et al., 1984; Rosenmann et al., 1985; Saadallah and Hulten, 1985; Chandley et al., 1986; Gabriel-Robez et al., 1986; Johannisson et al., 1987). In the pachytene spermatocytes of these translocation carriers, not only the asynapsed univalents, but the multivalents are also associated with sex chromosomes and the sex body, supporting the observation that at least small regions around the breakpoints are unsynapsed in multivalents. Later, interactions between the sex chromosome and translocated chromosome were clearly confirmed by immunohistochemistry with antibodies against the SC component in combination with fluorescence in situ hybridization technique (Olive-Bonet et al., 2005a). Notably, these autosomal–sex body associations are most likely involved with the etiology of male infertility in translocation carriers.
and females (Turner et al., 2005). Several lines of evidence also suggest that human females also possess a kind of surveillance system for synapsis. Female infertility is a common symptom in females with Turner syndrome and the 45, X karyotype, where X chromosome often remains unsynapsed. Examination of the ovaries of females with Turner syndrome reveals the depletion of pachytene oocytes (Speed, 1996). In Turner females with various structural abnormalities of the X chromosome, the size of the unsynapsed region appears to correlate with the severity of the sexual phenotype, suggesting that synapsis failure triggers apoptosis in oocytes (Ogata and Matsuo, 1995). These data suggest that the unsynapsed chromosome is detected by a quality control system even in oogenesis. According to this hypothesis, the surveillance system should respond to regional unsynapsis around the translocation breakpoints of the quadrivalent that leads to meiotic arrest both in males and females. This does not appear to reconcile with male-only infertility.

Here, there are two lines of conceptual thinking about the involvement of MSUC in synapsis failure that subsequently leads to the cell death of spermatocytes. One is that MSUC itself inactivates the indispensable autosomal genes, thereby leading to cell death. The inactivation of the involved autosomal segments might be enhanced by the presence of the sex body in pachytene spermatocytes, which can potentially explain the sex-specific difference in stringency of the system leading to male-only infertility. The alternative is MSCI failure, which means the derepression of transcription in sex chromosomal genes that leads to the subsequent apoptosis (Mahadevaiah et al., 2008). The unsynapsed region of the translocated chromosomes might inhibit the assembly of MSUC proteins in the sex body, although it is unclear whether a partially unsynapsed region is enough to impact this inhibitory effect. In addition, heterosynapsis between autosomes and sex chromosomes, which has been occasionally observed, might inhibit the silencing of the sex chromosomes. MSUC failure only happens in males, which might account for the sexual differences in the robustness of the checkpoints for synapsis in the presence of translocation.

**IDENTIFICATION OF SYNAPSIS CHECKPOINT PROTEIN IN MAMMALS**

To further investigate the mechanisms of the pachytene checkpoints at the molecular level, we searched for candidate genes that are essential to synapsis by examining the expression profiles of mouse genes expressed during the prophase of meiosis I (Kogo et al., 2012). Among the hundreds of candidate genes, we focused on Hormad1, the mammalian ortholog of yeast Hop1, by its potential function in the synapsis surveillance system. Hormad1 is a homology domain protein that is located on the SC in the unsynapsed region (Wojtasz et al., 2009; Fukuda et al., 2010). Recently, we and others have reported detailed analyses of Hormad1-deficient mice. Hormad1-deficient mice are infertile and demonstrate extensive failure of homologous pairing and synapsis in both males and females (Shin et al., 2010; Daniel et al., 2011; Kogo et al., 2012). In males, spermatogenesis arrests during the pachytene due to the severe synapsis failure, and all spermatogenic cells undergo stage IV apoptosis. Unexpectedly, Hormad1-deficient oocytes contain a normal number of oocytes, despite the extensive asynapsis, and consequently produce aneuploid oocytes that lead to subsequent fetal death in utero (Figures 3A,B). The failure to eliminate oocytes with extensive synapsis failure in the Hormad1-deficient ovary prompted the hypothesis that Hormad1 itself might be required for the mammalian pachytene checkpoint mechanism.

To exclude the effect of the DSB-dependent pachytene checkpoint and to independently analyze the checkpoint for synapsis, we utilized the Spo11-deficient mice that do not demonstrate meiotic DSB and thus do not demonstrate an active DSB-dependent pathway. Analysis of Hormad1/Spo11 double-knockout mice demonstrated that Hormad1 deficiency abrogates the massive oocyte loss that takes place in Spo11-deficient ovaries (Figures 3C,D). The double-mutant spermatocytes still undergo stage IV apoptosis, but they evade arrest at the zygotene/pachytene transition and the timing of cell death is slightly delayed (Kogo et al., unpublished data). These results raise speculation that the Hormad1 plays an important role in the DSB-independent pachytene checkpoint of both males and females, and possibly the surveillance for synapsis. Our present data thus highlight a novel DNA damage-independent synapsis checkpoint of mammalian meiosis.

Because spermatocytes still undergo apoptosis in male double-mutants, these mutants allowed us to analyze the male-specific backup pathway that is available for when the Hormad1-dependent pachytene checkpoint does not work. In Hormad1/Spo11 double-knockout males, derepression of sex chromosome genes was observed (Kogo et al., 2012). In the absence of Hormad1, spermatocytes fail to form the sex body. This might be due in part to the proteins required for heterochromatin formation, such as BRCA1, γH2AX, and ATR, that cannot be recruited to form the specialized chromatin domain if extensive asynapsis occurs, causing them to remain localized at DSB sites in unsynapsed regions (Mahadevaiah et al., 2008). Furthermore, in the absence of DSB, these components form the pseudo-sex body, as mentioned above, and do not assemble around the sex chromosomes. Consequently, transcriptional repression of the sex chromosome genes is impaired in male double-mutants, and this MSCI failure leads to spermatocyte apoptosis. Our data clearly demonstrate that one of these pathways acts as the male-specific surveillance system for synapsis failure.

Next, we set out to determine what mechanism triggers apoptosis at the Hormad1-dependent pachytene checkpoint for synapsis that operates both in males and females. One plausible idea is that the transcriptional silencing of autosomes via MSUC. As mentioned above, Spo11 deficiency induces the formation of the pseudo-sex body, within which the transcription of autosomes might be silenced, possibly leading to cell death. Hormad1 deficiency abrogates formation of the pseudo-sex body in Hormad1/Spo11 double-knockout females (Daniel et al., 2011; Kogo et al., 2012). Therefore, MSUC within the pseudo-sex body might be the cause of cell death in Spo11-deficient mice and, furthermore, it may be hormad1-dependent. On the other hand, another pathway that is not involved in MSUC also underlies the etiology of Hormad1-dependent cell death in synapsis failure (Kouznetsova et al., 2009). We found that Hormad1 is abundant at the unsynapsed chromosomes axes and highly phosphorylated in...
FIGURE 3 | Hormad1 deficiency abrogates massive apoptosis in Spo11 deficiency. (A) Representative image of a uterus. Hormad1-deficient females carry only a small number of implantation sites (right) compared with the control (left). The lower panel shows a representative image of a conceptus from a Hormad1-deficient female with a non-developing fetus (right). (B) Representative data of the first meiotic metaphase (MI) of wild-type (WT) and Hormad1-deficient oocytes with only a small number of bivalents (right). The lower panel shows a representative photograph of the second meiotic metaphase (MII) of Hormad1-deficient oocytes with some monovalents (right). (C) Hematoxylin and eosin staining of ovary sections from 20-day-old female mice. The ovaries from Spo11-deficient mice are small with only a small number of follicles, whereas Spo11/Hormad1 double-knockout mice demonstrate the same size and oocyte number as the wild-type levels. (D) The number of oocytes in 20-day-old mice ovaries. The c-kit positive oocytes are almost absent in the Spo11-deficient ovary, but are abundant in the Spo11/Hormad1 double-knockout ovaries (*P < 0.05).

Spo11-deficient mice (Kogo et al., 2012). We propose the hypothesis that extensive asynapsis activates the pachytene checkpoint signaling pathway via the phosphorylation of Hormad1, thereby leading to stage IV apoptosis in both males and females. A possible mechanism is illustrated in Figure 4A.

FUTURE PERSPECTIVES
To analyze the DSB-independent synapsis surveillance system independent of the DSB-dependent pachytene checkpoint, we had to utilize Spo11-knockout mice. However, Spo11 deficiency is a specific situation where most of the chromosomes are unsynapsed. It is still unknown whether the Hormad1-dependent pathway contributes to the consequence of partial unsynapsis due to translocation or other chromosomal abnormalities during normal meiotic prophase. Given the sexual dimorphism of the reproductive outcomes between male and female translocation carriers, the autosome–sex body association should provide useful clues regarding the molecular nature of the checkpoint mechanism for partial unsynapsis in spermatogenesis, regardless if this mechanism involves MSUC or MSCI failure (Figure 4B). If MSUC is the mechanism, Hormad1 might be an important part of the surveillance system, even during partial synapsis failure, because formation of the sex body is dependent on Hormad1, whereas in MSCI failure Hormad1 might not be essential. To date, there are a couple of studies on the etiology of infertile male mice with reciprocal translocation that have attempted to determine which mechanism is essential, but this is still an open question (Jaafar et al., 1993; Homolka et al., 2007). To evaluate these possibilities,
investigations on spermatogenesis in male infertile mice with reciprocal translocation under conditions of deficiency in MSUC component are needed.

On the other hand, it may also be possible that such autosome-sex body interactions do not necessarily lead to cell death. Infertile males with the Robertsonian translocation, whose spermatocytes show autosome-sex body association, mostly manifest as oligozoospermia, not azoospermia, suggesting that spermatocytes with partial unsynapsis could develop beyond the pachytene stage (Navarro et al., 1991). Indeed, mouse spermatocytes with the Robertsonian translocation manifest as partial synopsis failure with some evidence of MSUC, but can develop beyond the diploneme stage to undergo cell death at metaphase, possibly via the spindle assembly checkpoint (SAC; Eaker et al., 2001; Manterola et al., 2009). In addition to the reduced stringency of the oocyte SAC, attachment of a quadrivalent to the sex body might generate a problem and trigger male-specific effects at SAC during meiosis I (Oliver-Bonet et al., 2005a; Nagaoka et al., 2011). The involvement of SAC in the reproductive failure of translocation carriers warrants further investigations.

Another unsolved question is why some translocation carriers suffer from male infertility and others do not. The breakpoint location should have a considerable impact on MSUC and the reproductive outcome. Accordingly, Robertsonian translocations are expected to be less severe than reciprocal translocations because the breakpoints are located at the centromeric repeats that constitutes heterochromatin without any important genes, but in reality this is not true (Page et al., 1996). In non-Robertsonian reciprocal translocations, acrocentric chromosomes are frequently involved in cases of male infertility, but a relatively low rate of autosome-sex body association has been observed in reciprocal translocations not involved in acrocentric chromosomes (Chandley et al., 1986; Guichaoua et al., 1990). The close proximity of the sex body and nucleolus during pachytene stage might affect the susceptibility of MSUC to Robertsonian translocations (Kriebel et al., 1981; Tsutsumi et al., 2011).

In addition, there is one intriguing example that demonstrates whether infertility is dependent or independent of the location of the translocation breakpoints. In males with recurrent t(11;22)(q23;q11), only a subset of male translocation carriers are infertile, although they all possess almost identical breakpoints (Kurahashi et al., 2000). It can be hypothesized that the etiology behind synopsis failure-related infertility due to is multifactorial and that genetic factors govern the robustness of the synopsis checkpoint, thereby determining one’s susceptibility to infertility. Whole genome genotyping analysis of patients with reproductive failure and fertile controls might allow us to identify the risk factors and protective variants that might be located within the genes participating in the synopsis checkpoint (Aston and Carroll, 2009; Hu et al., 2011).

Together, MSUC and MSCI failure via autosome-sex body association might possibly render a subset of male translocation carriers infertile. Thus, this male-only infertility most likely contributes to the female predominance of the transmission of translocated chromosomes. The identification of Hormad1, which is the driver of the checkpoint system for extensive synopsis failure under conditions of Spo11 deficiency, might allow us to elucidate
of reproductive failure, infertility or RPL, due to chromosomal abnormalities. Kurahashi et al. Failure of synapsis and chromosomal disorders

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**REFERENCES**

Altmann, L. C., and Carroll, D. T. 2005. Genome-wide study of single-nucleotide polymorphisms associated with aspermia and severe oligozoospermia. J. Androl. 26, 714–722.

Barnes, M. G., and Gondek, J. A. 2001. Chromatin dynamics in the male meiotic prophase. Cytogenet. Genome Res. 103, 225–234.

Barclay, M., Maheshwari, S. D. G practicing medicine yields distinct responses despite elimination at an essential developmental stage. Mol. Cell Biol. 25, 7213–7223.

Baudat, F., Buard, J., Barthès, P., Grey, C., and Fledel-Baarends, W. M., and Grootegoed, J. A. 2009. Histone H3 lysine 4 trimethylation and transcriptional activity at the pachytene stage. J. Cell Sci. 122, 3684–3694.

Baudot, H., Mori, T., Nishiyama, S., Ito, T. (2009). Genome-wide study for sex-body formation, and Spo11 se-post-divisional recombination defects in mouse spermatogenesis. Jpn. J. Cytogenet. Genome Res. 51, 225–235.

Baudot, F., Buard, J., Grey, C., Fledel-Baarends, W. M., and Grootegoed, J. A. 2009. Histone H3 lysine 4 trimethylation and transcriptional activity at the pachytene stage. J. Cell Sci. 122, 3684–3694.

Bolum, H., Mori, T., Nishiyama, S., Ito, T. (2009). Genome-wide study for sex-body formation, and Spo11 se-post-divisional recombination defects in mouse spermatogenesis. Jpn. J. Cytogenet. Genome Res. 51, 225–235.
Kobielu, B., Miró, C., Hartung, M., Jean, P., and Stahl, A. (1981). Sex vesicle-associated nuclear organizers in mouse spermatozoa: localization, structure, and function. Cytogenet. Cell Genet. 31, 47–57.

Koga, K., Kose-Sugiyama, H., Tamada, K., Bulet, H., Tsutsumi, M., Ohno, T., Inagaki, H., Tsuchiya, M., Toda, T., and Kurahashi, H. (2010). Screening of genes involved in chromosome segregation during meiosis I toward the identification of genes responsible for infertility in humans. J. Hum. Genet. 55, 259–259.

Koga, H., Tsutsumi, M., Ohno, T., Inagaki, H., Tsuchiya, M., Ohye, T., and Kurahashi, H. (2011). TERM syndrome and female sex chromosome aberrations: deduction of the principal factors involved in the development of clinical features. Hum. Genet. 95, 607–629.

Ohno, T., Inagaki, H., Koga, H., Tsutsumi, M., Kato, T., Tong, M., Miyazaki, M. V., Madare, S., Zakus, E. H., Emanuel, B. S., and Kurahashi, H. (2010). Partial origin of the de novo constitutive t(1;22)(q11;q11). J. Hum. Genet. 55, 293–299.

Kurahashi, H., Tsuchiya, M., and Kurahashi, H. (2010). Screen- ing of genes involved in chromosome segregation during meiosis I: toward the identification of genes responsible for infertility in humans. J. Hum. Genet. 55, 259–259.

Kobielu, B., Miró, C., Hartung, M., Jean, P., and Stahl, A. (1981). Sex vesicle-associated nuclear organizers in mouse spermatozoa: localization, structure, and function. Cytogenet. Cell Genet. 31, 47–57.

Koga, K., Kose-Sugiyama, H., Tamada, K., Bulet, H., Tsutsumi, M., Ohno, T., Inagaki, H., Tsuchiya, M., Toda, T., and Kurahashi, H. (2010). Screening of genes involved in chromosome segregation during meiosis I toward the identification of genes responsible for infertility in humans. J. Hum. Genet. 55, 259–259.

Koga, H., Tsutsumi, M., Ohno, T., Inagaki, H., Tsuchiya, M., Ohye, T., and Kurahashi, H. (2011). TERM syndrome and female sex chromosome aberrations: deduction of the principal factors involved in the development of clinical features. Hum. Genet. 95, 607–629.

Ohno, T., Inagaki, H., Koga, H., Tsutsumi, M., Kato, T., Tong, M., Miyazaki, M. V., Madare, S., Zakus, E. H., Emanuel, B. S., and Kurahashi, H. (2010). Partial origin of the de novo constitutive t(1;22)(q11;q11). J. Hum. Genet. 55, 293–299.

Kurahashi, H., Tsuchiya, M., and Kurahashi, H. (2010). Screen- ing of genes involved in chromosome segregation during meiosis I: toward the identification of genes responsible for infertility in humans. J. Hum. Genet. 55, 259–259.

Kobielu, B., Miró, C., Hartung, M., Jean, P., and Stahl, A. (1981). Sex vesicle-associated nuclear organizers in mouse spermatozoa: localization, structure, and function. Cytogenet. Cell Genet. 31, 47–57.

Koga, K., Kose-Sugiyama, H., Tamada, K., Bulet, H., Tsutsumi, M., Ohno, T., Inagaki, H., Tsuchiya, M., Toda, T., and Kurahashi, H. (2010). Screening of genes involved in chromosome segregation during meiosis I toward the identification of genes responsible for infertility in humans. J. Hum. Genet. 55, 259–259.

Koga, H., Tsutsumi, M., Ohno, T., Inagaki, H., Tsuchiya, M., Ohye, T., and Kurahashi, H. (2011). TERM syndrome and female sex chromosome aberrations: deduction of the principal factors involved in the development of clinical features. Hum. Genet. 95, 607–629.

Ohno, T., Inagaki, H., Koga, H., Tsutsumi, M., Kato, T., Tong, M., Miyazaki, M. V., Madare, S., Zakus, E. H., Emanuel, B. S., and Kurahashi, H. (2010). Partial origin of the de novo constitutive t(1;22)(q11;q11). J. Hum. Genet. 55, 293–299.

Kurahashi, H., Tsuchiya, M., and Kurahashi, H. (2010). Screen- ing of genes involved in chromosome segregation during meiosis I: toward the identification of genes responsible for infertility in humans. J. Hum. Genet. 55, 259–259.

Kobielu, B., Miró, C., Hartung, M., Jean, P., and Stahl, A. (1981). Sex vesicle-associated nuclear organizers in mouse spermatozoa: localization, structure, and function. Cytogenet. Cell Genet. 31, 47–57.

Koga, K., Kose-Sugiyama, H., Tamada, K., Bulet, H., Tsutsumi, M., Ohno, T., Inagaki, H., Tsuchiya, M., Toda, T., and Kurahashi, H. (2010). Screening of genes involved in chromosome segregation during meiosis I toward the identification of genes responsible for infertility in humans. J. Hum. Genet. 55, 259–259.

Koga, H., Tsutsumi, M., Ohno, T., Inagaki, H., Tsuchiya, M., Ohye, T., and Kurahashi, H. (2011). TERM syndrome and female sex chromosome aberrations: deduction of the principal factors involved in the development of clinical features. Hum. Genet. 95, 607–629.

Ohno, T., Inagaki, H., Koga, H., Tsutsumi, M., Kato, T., Tong, M., Miyazaki, M. V., Madare, S., Zakus, E. H., Emanuel, B. S., and Kurahashi, H. (2010). Partial origin of the de novo constitutive t(1;22)(q11;q11). J. Hum. Genet. 55, 293–299.

Kurahashi, H., Tsuchiya, M., and Kurahashi, H. (2010). Screen-ing of genes involved in chromosome segregation during meiosis I: toward the identification of genes responsible for infertility in humans. J. Hum. Genet. 55, 259–259.
and leads to substantial postmeiotic repression in spermatids. Dev. Cell 10, 521–529.

Turner, J. M., Mahadevaiah, S. K., Fernández-Capetillo, O., Nissen-Zwing, A., Xu, X., Deng, C. X., and Burgoyne, P. S. (2005). Silencing of unpaired meiotic chromosomes in the mouse. Nat. Genet. 37, 41–47.

Wojtasz, L., David, R., Borg, J., Belcaro-Viles, E., Xu, H., Boonman, V., Edelman, C. R., Goeke, H. J., Jasin, M., Kerney, S., McKay, M. I., and Toid, A. (2009). Mouse HORMAD1 and HORMAD2, two conserved meiotic chromosomal proteins, are depleted from synapsed chromosome axes with the help of TRIP13 AAA ATPase. PLoS Genet. 5, e1000702. doi: 10.1371/journal.pgen.1000702

Yan, W., and McCarey, J. R. (2009). Sex chromosome inactivation in the male. Epigenetics 4, 452–458.

Yang, F., De La Fuente, R., Liu, N. A., Brummer, C., McLaughlin, K. L., and Wang, P. J. (2006). Mouse SCP3 is required for synaptonemal complex assembly and chromosomal synapsis during male meiosis. J. Cell Biol. 175, 497–507.

Yoshida, K., Kendol, G., Matsuoka, Y., Hata, T., Nishimura, E., and Merker, T. (1998). The mouse Rad51 like gene Dmc1 is required for homologous chromosome synap- sis during meiosis. Mol. Cell 1, 707–718.

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