Anomalous X chromosome inactivation: the link between female zygotes, monozygotic twinning, and neural tube defects?

SIR,

Two hypotheses have recently been offered suggesting that anomalous X inactivation is causally connected with monozygotic (MZ) twinning\(^1\) and neural tube defects\(^2\) respectively. The authors of the first of these papers were rather hesitant in their presentation and critical of their own hypothesis. I want here to offer grounds for supposing that criticism to be misguided, and to note the wealth of hitherto unexplained data which would become comprehensible if both these hypotheses were true.

The criticism concerns the sex ratio of MZ twins. Burn et al\(^4\) noted that if anomalous X inactivation were to cause MZ twins, then MZ twins should contain an excess of females, but they suggested that (except for conjoined pairs) MZ twins do not contain such an excess. However, there is indirect\(^3\) and direct\(^4\) evidence for such a female excess, this excess being greater the later the twins are formed.

Boklage\(^5\) suggested that the probability distribution of MZ twinning events in developmental time is roughly normal with a mean of 4-7 days (after fertilisation) and a standard deviation of 1-3 days. It is thought that the placenta of MZ twins is a guide to the time of their formation, dichorionic pairs being first, then monochorionic diamniotic pairs, then monoamniotic pairs, and finally conjoined pairs. If this is correct, then using Boklage’s estimates of the percentages of these placental types among MZ twins, one may estimate the time intervals during which the various placental types are initiated. These values are given in the table. This table also cites the sex of MZ pairs by placentaion in my paper\(^4\) and (based on these sex ratios) estimates of the percentages of such twins initiated by anomalous X inactivation.

I know of no direct data on the timing of X inactivation in human embryos, but Monk\(^7\) suggests a date of around five days for the relevant tissues in the mouse, that is, about eight to 10 days in the human being.\(^8\) Bearing in mind the elasticity of all these estimates, it seems that in a few cases (mainly where the MZ twinning event occurs late) this MZ event and the X inactivation may roughly coincide. If the one were to cause the other, there would be a very attractive explanation of the decline in sex ratio of MZ twins with the developmental time at which they are initiated.

As noted above, if this hypothesis and that of Hall\(^2\) (that anomalous X inactivation is sometimes responsible for neural tube defects) were both true, then explanations would become available for a number of established but unexplained features of the epidemiology of neural tube defects including the following.

(1) The excess of anencephaly in MZ but not DZ twins.\(^9\)

(2) The increase of risk of anencephaly with the time

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**TABLE Monozygotic twins by placental class. Columns a–c: their sexes and sex ratio (data cited by James\(^4\)). Column d: their relative percentages among all MZ twins (estimated by Boklage\(^5\)). Column e: the days on which they are initiated (estimated here from the data of Boklage\(^5\)). Column f: the percentage of each which is estimated here to be caused by anomalous X inactivation.**

<table>
<thead>
<tr>
<th>Placental class</th>
<th>a</th>
<th>b</th>
<th>c</th>
<th>d</th>
<th>e</th>
<th>f</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Male</td>
<td>Female</td>
<td>Sex ratio</td>
<td>Relative percentages</td>
<td>Days of initiation</td>
<td>Percentage estimated caused by anomalous X inactivation</td>
</tr>
<tr>
<td>Dichorionic</td>
<td>76</td>
<td>57</td>
<td>0.571</td>
<td>31-1</td>
<td>-4</td>
<td>Nil</td>
</tr>
<tr>
<td>Monochorionic diamniotic</td>
<td>694</td>
<td>717</td>
<td>0.492</td>
<td>65-5</td>
<td>4-7</td>
<td>4-5</td>
</tr>
<tr>
<td>Monochorionic monoamniotic</td>
<td>126</td>
<td>177</td>
<td>0.416</td>
<td>2-4</td>
<td>7-7½</td>
<td>19</td>
</tr>
<tr>
<td>Conjoined</td>
<td>22</td>
<td>74</td>
<td>0.229</td>
<td>1-0</td>
<td>7½+</td>
<td>More than 50</td>
</tr>
</tbody>
</table>

(1) The twins called ‘monochorionic diamniotic’ contain a few (of the order of 20) monochorionic monoamniotic pairs. (2) The twins called ‘monochorionic monoamniotic’ exclude conjoined pairs. (3) Let us suppose that no dichorionic MZ twins are caused by anomalous X inactivation (being formed before X inactivation normally occurs). Accordingly their true sex ratio may be supposed to be 0.514, a typical sex ratio for liveborn Caucasian singletons (and a value not significantly different from the direct data cited here on dichorionic MZ twins). Then the weighted mean of the sex ratios estimated for MZ twins of the various placental classes in the table is 0.494. This is close to the value of 0.492 estimated by indirect means and thus provides some measure of support for both the direct and indirect estimates cited in the text.
at which MZ twins are initiated. Anencephaly is least common in dichorionic MZ pairs and most common in conjoined and other monoamniotic pairs.\textsuperscript{10} (It is assumed that the closer in time that an anomalous event occurs to a developmental process, the more likely that process is to be affected deleteriously.)

(3) The raised concordance rate for anencephaly in MZ twins but not DZ twins, bearing in mind the risk.\textsuperscript{11}

(4) The low sex ratio of spina bifida cases that occur in twins.\textsuperscript{12}

It has been suggested that the deleterious effect of anomalous X inactivation on neural tube development is mediated by environmental factors.\textsuperscript{2} This would explain the fact that when anencephaly rates vary with a factor (for example, social class, season, region, time, maternal age, parity, race) the sex ratio of cases varies too: when rates are high, sex ratios are low, suggesting that environmental cofactors (together with anomalous X inactivation) are responsible for varying proportions of female cases.

The closure of the neural tube is a directional process occurring in general a few days after X inactivation and MZ twinning events. It was suggested above that the closer in time that an anomalous event occurs before a developmental process, the more likely that process is to be affected deleteriously. If this were so, then anencephaly (being initiated closer in time than spina bifida to X inactivation) would be expected to be more frequently caused by anomalous X inactivation. This would explain (1) the generally lower sex ratio of anencephaly cases than spina bifida cases,\textsuperscript{12,13} (2) the fact that anencephaly is more closely related to MZ twinning than is spina bifida,\textsuperscript{10} and (3) the rather general lack of association between the sex ratio of spina bifida cases (in contrast with anencephaly cases) and incidence rates.\textsuperscript{12}

I am grateful to Dr Anne McLaren FRS for help.

WILLIAM H JAMES

MRC Mammalian Development Unit,
Wolfson House,
University College London,
4 Stephenson Way,
London NW1 2HE.

References


Duchenne muscular dystrophy in one of monozygotic twin girls

Sir,

Burn et al\textsuperscript{1} refer to two cases of pseudohypertrophic muscular dystrophy cases and another of Christmas disease in one of monozygotic (MZ) 46,XX twin girls and add a further MZ 46,XX twin pair discordant for Duchenne muscular dystrophy. In this case they showed by somatic cell hybridisation that in one twin girl only the maternal X chromosome was active, whereas in the other only the paternal one was active. Other MZ twins, discordant for colour blindness\textsuperscript{2,3} or G6PD deficiency,\textsuperscript{4} also support their contention that such discordance does not arise by chance after random X inactivation but as a result of an unusual pattern of unequal X chromosome inactivation: one female in whom (nearly?) all cells express the X chromosome with the related gene, and the other in whom this gene is inactivated. Extending the hypothesis of 'unequal lyonisation', they state that the abnormal segregation of cells might actually have caused MZ twinning. This extension, however, does not explain the not infrequent observation of MZ twins discordant for chromosome abnormalities, that is, heterokaryotypic twinning and, as they agree, the predicted preponderance of female MZ twins is not substantiated in MZ twin samples.

The general phenomenon of structural aberrations of the X chromosome being associated with late replication and inactivation in Turner patients, currently explained by 'preferential' inactivation, led me to wonder if this unusual phenomenon might