

Fetal Behavior and Heart Rate in Twin Pregnancy: A Review

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Fetal movements and fetal heart rate (FHR) are well-established markers of fetal well-being and maturation of the fetal central nervous system. The purpose of this paper is to review and discuss the available knowledge on fetal movements and heart rate patterns in twin pregnancies. There is some evidence for an association or similarity in fetal movement incidences or FHR patterns between both members of twin pairs. However, the temporal occurrence of these patterns seems to be for the most part asynchronous, especially when stricter criteria are used to define synchrony. The available data suggest that fetal behavior is largely independent of sex combination, fetal position, and presentation. Conversely, chorionicity appears to have some influence on fetal behavior, mainly before 30 weeks of gestation. There is preliminary evidence for the continuity of inter-individual differences in fetal activity and FHR patterns over pregnancy. Comparisons between studies are limited by large methodological differences and absence of uniform concepts and definitions. Future studies with high methodological quality are needed to provide a more comprehensive knowledge of normal fetal behavior in twin pregnancy.

■ **Keywords:** twin pregnancy, fetal movement, fetal heart rate, fetal behavior

Twins have been the subject of great interest since ancient times. However, it was not until the second half of the 19th century that twins ceased to be regarded as an interesting phenomenon (natural wonder) and an obstetrical challenge. This attitude toward twins changed after the appearance in of Galton's study in 1876: 'The history of twins as a criterion of the relative powers of nature and nurture'. Galton was not only the first to postulate the existence of two types of twins (zygosity) but also laid the foundation of the (behavioral) genetic research carried out until now.

Because monozygotic twins are genetically identical, every difference between them is believed to be due to the environment (nurture). On the other hand, dizygotic twins share a similar genetic background as brothers and sisters, but like monozygotic twins they are of the same age, share the same uterus at the same time, and to a certain extent develop under similar circumstances. By quantifying similarities and differences in various outcomes between monozygotic and dizygotic twins, one may start to determine the relative contributions of heredity (nature) and environmental factors (nurture) during human development (Plomin & Asbury, 2005). Thus, prenatal observation of fetal movements simultaneously performed in both halves of monozygotic or dizygotic twins could be of great

importance for behavioral genetics research, developmental neurobiology, and for the evaluation of the condition of fetuses, especially with respect to interfetal growth discordances and in cases of twin–twin transfusion syndrome (TTTS).

Besides genetic and environmental main effects, several forms of gene–environment interplay have been identified. Overall, they indicate that the influence of genes and environments on human development is not completely independent. The findings on epigenetic effects, for example, demonstrate that gene expression is influenced by environmental factors. Environmental risk exposure has also been found to be influenced by genetic factors (gene–environment correlations; Rutter et al., 2006). During ontogeny, there is a continuous interaction between genetic and environmental factors, which basically starts at

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the zygotic stage. The study of prenatal behavior in twin pregnancies starting in early pregnancy may give insight into the causation of fetal behavior and factors that possibly play a role, such as maternal factors (hormones, heart rate and bowel sounds, stress), blood flow to the uterus, fetal position, and inter-twin contacts (physical or humoral, e.g., via a third circulation).

Today, the scientific study of twins represents a preferred method in behavioral genetics, developmental studies, matched-pair experimental designs and epidemiology. At the same time, important applications are found in an increasing number of areas, particularly in psychology and psychiatry, medical genetics, reproductive biology and obstetrics, and neonatology. For instance, advances in ultrasound monitoring have facilitated a shift of developmental psychology frontiers from the neonatal to fetal period. Knowledge about the origins and development of individual differences, as well as the factors that may influence this process, can now be expanded with the study of behavior throughout pregnancy.

Fetal movements and fetal heart rate (FHR) have been extensively analyzed to describe normal development over the course of pregnancy, especially in singleton pregnancies. These variables are considered indirect measures of the fetal central nervous system (CNS) functioning (de Vries & Fong, 2006). Fetal movements follow a developmental course over pregnancy (de Vries et al., 1988; Roodenburg et al., 1991; ten Hof et al., 2002) and abnormal fetal movements have been observed in fetuses with brain dysfunction (de Vries & Fong, 2007). Similarly, abnormal heart rate patterns are suggestive of fetal distress. The association of fetal movements with heart rate patterns into specific combinations characterizes fetal behavioral (sleep–wake) states that resemble those described in the neonate. Disturbed development of fetal sleep organization has, for instance, been found in fetuses of women with type-1 diabetes (Mulder et al., 1987) and in fetuses exposed to antidepressants (Mulder et al., 2011).

Fetal Movements and Fetal Heart Rate Research

General Considerations and Definitions

Throughout this review, the original terminology used by various researchers, such as fetal ‘behavior’, ‘movements’, ‘motility’, ‘motor behavior’ or ‘motor activity’, ‘reactive’, or ‘evoked’ movements, is maintained. These terms are often employed interchangeably.

Fetal behavior has been defined as ‘any observable action or reaction (to an external stimulus)’ (Hepper, 1996, p. 145). ‘Fetal behavior’ and ‘fetal movements’ are employed in some studies as synonyms (de Vries et al., 1982, 1985; Piontelli et al., 1999). In the literature, the assessment of fetal behavior has also been referred to as the simultaneous assessment of fetal activity and FHR patterns (Nijhuis, 2003).

However, none of the reviewed studies adopted this last definition.

Spontaneous fetal movements have been described as non-reflexive, autonomous movements originating in the fetus itself whereas the evoked fetal movements were thought to be caused by a variety of stimuli (in the fetal environment), being reflex in origin (Goodlin & Lowe, 1974; Graves, 1980; Preyer, 1882/1888).

In the next section, we provide a brief overview of what is known about normal fetal functional development in singletons, followed by a review of studies that have focused on fetal movements and heart rate in twin pregnancy.

Fetal Movements

Research on singletons has shown that normal fetal movement is characterized by wide interfetal variations in incidence of fetal movement patterns (sideways bending, general movement, startle, hiccup, breathing movement, stretch, yawning, isolated arm or leg movements, hand–face contact, jaw opening, sucking and swallowing, and head retroflexion, rotation, and antelexion) from its early appearance to the end of gestation. It is striking that all fetal movement patterns have their onset before 14 weeks of gestation, although their incidences change over pregnancy (de Vries et al., 1982; Roodenburg et al., 1991).

In twins, the onset and incidence of specific movement patterns has been less investigated than in singletons. In contrast, researchers were more interested in the overall activity level of twins as compared with singletons in the synchrony of fetal movements and in the rate of spontaneous and evoked fetal movements. Few studies have examined specific variables of twin pregnancy that may have an association with fetal movements, such as zygosity, chorionicity, sex combination (male–male, male–female, female–female), and other variables such as fetal position (longitudinal or transverse lie) or presentation (cephalic, breech, or transverse).

Ultrasound observations of fetal movements provide the most accurate means to analyze fetal activity, as an experienced observer is able to differentiate between spontaneous, evoked, and passive movements of twins. Hence, we will provide an overview of studies that employed this technique to study fetal movements in twins.

Quantitative analysis of fetal movements of twins was performed in eight studies using two-dimensional (2D) ultrasound (Arabin et al., 1996; Mulder et al., 2004, 2012; Piontelli et al., 1997, 1999; Sadovsky et al., 1987; Samueloff et al., 1991; Zimmer et al., 1988) and more recently with 4D ultrasound (Degani et al., 2009; Hata et al., 2011).

The incidence of spontaneous movement patterns studied in the first half of twin pregnancies has been found to be similar to that in singletons (Piontelli et al., 1997). However, others recently found that twins have a lower incidence of general movements than singletons over pregnancy, but

TABLE 1
Definitions of Synchrony/Simultaneity in Fetal Movements (FM)
and Fetal Heart Rate (FHR) Adopted in Twin Studies

Definitions of synchrony/simultaneity	Studies
A movement/FHR acceleration of one twin is followed by a movement/FHR acceleration of the co-twin within 15 seconds.	Gallagher et al. (1992) (FHR, FM) Sherer et al. (1990, 1991, 1992) (FHR)
Movements/FHR accelerations of both twins start exactly at the same time.	Mulder et al. (2012) (FM) Sadovsky et al. (1987) (FM) Zimmer et al. (1988) (FM)

more breathing movements in the last trimester (Mulder et al., 2012).

The incidence of several fetal movement patterns seems to be fairly correlated within twin pairs over pregnancy, ranging from 0.30 for general movements to 0.72 for breathing movements (Mulder et al., 2012). Stronger associations were found for all studied fetal movement patterns before 30 weeks of gestation.

The temporal coincidence of fetal movements in both twins has been investigated in several studies. However, the adopted definition of synchrony/simultaneity varied between studies (see Table 1). Fetal movements or FHR accelerations were defined as synchronous/simultaneous when a fetal movement or an FHR acceleration of one twin was seen at the same time as in the co-twin up to 15 seconds apart. In this review, we will focus only on the results of studies in which synchrony was defined as the immediate start of fetal movements, heart rate patterns, or the association between both.

With 2D ultrasound, the reported rate of simultaneous movements in both twins studied in the first half of pregnancy was 5% (Samueloff et al., 1991), 24–26% in the last trimester of pregnancy (Sadovsky et al., 1987; Zimmer et al., 1988), and 10–80% throughout pregnancy, using different window lengths to define simultaneity (Mulder et al., 2012). Analysis of fetal movements revealed significant increase in breathing movement synchrony over pregnancy within twin pairs, while no gestational trend was found for general movements' synchrony (Mulder et al., 2012).

Spontaneous movements seem to be more frequent than evoked movements throughout twin gestation. As stated previously, spontaneous movements were defined as those originating in the fetus itself, while evoked movements were believed to be caused by a variety of stimuli. In the first half of pregnancy, spontaneous movements represent 88% of overall activity in twin pregnancies at 13 weeks and 71% at 20–22 weeks (Piontelli et al., 1997). A higher mean rate of spontaneous fetal movements (95%) was reported in a cross-sectional study that included twin pregnancies at 10 to 22 weeks (Samueloff et al., 1991). The same research team reported a mean proportion of 76% of spontaneous fetal movements in the second half of pregnancy (Sadovsky et al., 1987).

Based on a limited number of studies, the occurrence of fetal movement patterns seems largely independent of variables such as zygosity, chorionicity, sex combination, position, and presentation. No difference in fetal movements' synchrony or incidence of most fetal movement patterns has been found according to zygosity and chorionicity (Arabin et al., 1995; Hata et al., 2011; Mulder et al., 2012; Piontelli et al., 1997, 1999; Zimmer et al., 1988). Similar levels of spontaneous (Piontelli et al., 1999) and evoked fetal movements were found in monochorionic (MC) and dichorionic (DC) twins from 12 to 22 weeks of pregnancy (Hata et al., 2011; Piontelli et al., 1997, 1999). However, in one study MC twins showed a more substantial decline in spontaneous activity than opposite-sex DC twin pairs after 15 weeks (Piontelli et al., 1999). Earlier findings that pointed to an association between fetal position and breathing movement incidence in late pregnancy (Zimmer et al., 1988) were not confirmed by a recent longitudinal study (Mulder et al., 2012). No difference in total fetal activity (amount of time the fetus breathed and moved) has been found to be associated with fetal presentation (Zimmer et al., 1988) or position (Mulder et al., 2012).

Inter-Twin Contacts

The existence of contacts between both twins during pregnancy has been somewhat controversial in the late 1980s and early 1990s. Sadovsky et al. (1987) and Samueloff et al. (1991) on the basis of their ultrasound observations in each trimester of pregnancy rejected the possibility that movements of one twin stimulated the co-twin. However, Sherer et al. (1990, 1992) suggested that 'tactile communication' between twin fetuses could explain coinciding fetal heart accelerations during non-stress testing.

These contacts have been described in a small number of ultrasound studies carried out from the mid-1990s to the present time either by using conventional 2D (Arabin et al., 1996; Piontelli et al., 1997) or 4D ultrasound (Hata et al., 2011; Sasaki et al., 2010). A consistent finding across these studies is that inter-twin contacts seem to occur earlier in MC than in DC twins.

Arabin et al. (1996) investigated the onset and development of inter-twin contacts until 16 weeks of pregnancy and classified them according to their duration, reaction of the co-twin (present or absent), velocity, and body parts involved. The first contacts with a reaction of the co-twin were first observed in MC twins around 8 weeks (Arabin et al., 1996; Piontelli et al., 1997). Inter-twin contacts were found more frequently in MC than DC twins until 16 weeks of pregnancy. The onset of complex contacts (lasting >5 seconds) was observed earlier in the same-sex female twins, while an increased frequency of these contacts was found in the same-sex male pairs at 8–16 weeks (Arabin et al., 1996). Piontelli et al. (1997) observed steady increase in time spent in evoked movements from 11–13 weeks to 18–19 weeks followed by a plateau at 20–22 weeks. While no

specific developmental trend is evoked, movements were identified by these authors, some movement patterns were observed more frequently at particular gestational periods. A lack of response to an unequivocal contact was noted at all studied ages from 10–22 weeks. Using 4D ultrasound, Hata et al. (2011) found that at 12–13 weeks most inter-twin contacts do not elicit a reaction by the co-twin, even though a wide variation in reaction rates was reported. In addition, no significant difference in the rate of reaction was found according to chorionicity. Consistent with the results obtained in previous studies using 2D ultrasound, MC twins showed significantly more contacts than DC twins at the end of the first trimester of pregnancy (10–11 weeks of gestation) studied by 4D ultrasound (Sasaki et al., 2010). However, no differences were found between both groups at 12–13 weeks, as these contacts increased significantly in DC twins whereas contacts in MC twins remained stable.

Inter-twin contacts assessed by ultrasound in the last trimester of pregnancy have not been described yet, probably due to limitations on simultaneous monitoring of the entire body of both twins.

Three studies investigated the existence of a ‘dominant twin’ in a twin pair during pregnancy (Piontelli et al., 1997, 1999; Sherer et al., 1992), although the definitions of ‘dominance’ were different. Sherer et al. (1992) analyzed repeated non-stress FHR tracings of twin pairs of at least 30 weeks’ gestation to investigate whether inter-twin contacts were consistently initiated by the same twin (‘dominant twin’). It was assumed that the presence of FHR acceleration in both twins occurring within a 15-second period was caused by a previous inter-twin contact. The results showed that the initiation of inter-twin contacts was not systematically associated to one of the twins. Similarly, Mulder et al. (2012) found no evidence of a ‘dominant twin’ as demonstrated by higher fetal activity than the co-twin throughout pregnancy. In contrast, in another study using 2D ultrasound, dominance by one of the twins was found to be consistent from 10 to 22 weeks of gestation, as indicated by higher overall spontaneous activity level (Piontelli et al., 1999). In addition, it was noted that most of the ‘dominant twins’ in each pair had significantly lower evoked movements than their co-twins. Although the authors interpret this finding as the possible result of lower fetal stimuli generated by the less active twin, the number of contacts initiated by each twin was not reported (Piontelli et al., 1999). The contradictory results of these studies are probably explained by the adoption of different definitions of ‘dominance’, and diverse methodologies (ultrasound vs. electronic fetal monitoring) and study periods during pregnancy.

Fetal Heart Rate

Studies on normal, low-risk singletons have revealed developmental trends in FHR and its variation: With advancing maturation there is gradual decline in resting FHR and increase in FHR variability and FHR accelerations (Nijhuis

et al., 1998; Ohel et al., 1985; Serra et al., 2009; ten Hof et al., 1999). These characteristics are recognizable early in pregnancy and follow a continuous gestational trend (Serra et al., 2009). In addition, there is an evidence of diurnal variation in mean FHR and its variability from mid-gestation onwards (Visser et al., 1982; Vries et al., 1987).

Twins and singletons display a similar frequency of heart rate accelerations from 28 weeks onwards (Ohel et al., 1985).

Most of the research on FHR in twins has focused on the intra-pair coincidence of heart rate patterns and accelerations as a potential marker of zygosity type or evoked fetal movements resulting from inter-twin stimulation.

There is some evidence for the association between chorionicity and synchrony of accelerations. Twins with synchronous accelerations were more likely to be MC or have fused DC placentas and smaller intra-pair birth weight differences than asynchronous twin pairs (Devoe, 1985; Devoe & Azor, 1981). Overall, 58% of FHR patterns were classified as synchronous, although no specific definition of synchrony was provided (Devoe & Azor, 1981). In another study, Sherer et al. (1992) found that less than 10% of all accelerations were synchronous. Synchronous heart rate accelerations have also been noted immediately after vibroacoustic stimulation (Sherer et al., 1991).

Similar patterns in diurnal variation in FHR of twins have been consistently found regardless of chorionicity (Lunshof et al., 1997; Maeda et al., 2006; Muro et al., 2001). A significant intra-pair correlation in diurnal rhythms of FHR baseline was found (Maeda et al., 2006; Muro et al., 2001). In contrast, no intra-pair coincidence of sustained fetal tachycardia was noted (Maeda et al., 2006; Muro et al., 2001). Sustained fetal tachycardia is a characteristic of FHR pattern D, the specific heart rate pattern of the fetal behavioral state 4F described by Nijhuis et al. (1982) that corresponds to active awake state. These findings were drawn from the studies that included uncomplicated (Maeda et al., 2006; Muro et al., 2001) and mildly complicated twin pregnancies (mild maternal hypertension, discordant fetal growth; Lunshof et al., 1997).

Nonlinear analysis of heart rate variability revealed smaller intra-pair differences in nonlinear properties of FHR in MC than in DC twins before 30 weeks (Shiba et al., 2008). However, from 30 to 36 weeks, MC twins seem to become increasingly different in FHR-regulating mechanisms. It has been suggested that this could be explained by an increasing influence of maternal and environmental factors over pregnancy. Nevertheless, the results also indicate that both MC and DC twin fetuses followed a similar trend in the heart rate dynamics, which is progressively more chaotic with increasing gestational age.

Association Between FHR and Fetal Movements

With advancing gestation, the fetal quiescence and activity periods are progressively extended and some linkage between heart rate, body movement, and eye movements

occurs between 25 and 32 weeks in singleton pregnancies (Drogtróp et al., 1990; Visser et al., 1987). The combination of the three state variables into the well-defined behavioral states that resemble those in the neonate can be recognized at 36–38 weeks of gestation (Nijhuis et al., 1982).

None of the few twin studies that examined association between FHR patterns and fetal movements have applied the strict criteria proposed by Nijhuis et al. (1982) to define behavioral states, which would require the use of two FHR monitors and four ultrasound transducers for body and eye movements. The simultaneous observation of FHR and fetal movements has mostly been studied in twins by means of a fetal actocardiograph. To assess the effects of maternal betamethasone administration in twin pregnancy, FHR and fetal movements have been collected simultaneously with the use of four ultrasound devices (Mulder et al., 2004). However, these variables were described separately and their association has not yet been reported.

Gallagher et al. (1992) examined fetal behavior patterns of twins, a modification of fetal behavioral states that relies on the FHR and fetal movement monitoring provided by actocardiograph without the use of permanent ultrasonography. Ninety-five percent of the fetal behavior patterns (sleep or awake states) were synchronous. Interestingly, the same-sex twins were found to have greater synchrony in fetal behavior patterns than opposite-sex twins, whereas no significant differences were found between MC and DC twins (100% and 92% of the time respectively).

Arabin et al. (1993) classified FHR and fetal movement patterns occurring at the same time in twin pairs as ‘concordant’ or ‘discordant’, using a 3-minute window. Fetal behavioral patterns were categorized as ‘occasionally’ or ‘continuously’ ‘concordant’ or ‘discordant spontaneous behavior’. No systematic changes were identified in the distribution of all patterns from 26 to 36 weeks (Arabin & van Eyck, 2006). According to the authors, twins exhibit a higher percentage of active periods (mean 80%) than singletons, probably caused by inter-twin contacts.

Stability of Individual Differences

Intra-fetal consistency in fetal movements in singletons has been shown to be weak during pregnancy (de Vries et al., 1988; ten Hof et al., 2002), whereas less intra-fetal variation has been found for FHR parameters (Visser et al., 1982) and fetal behavioral states (Groome et al., 1995). A number of studies on singletons have documented a continuity of inter-individual differences in FHR and movement patterns observed in the second half of gestation and infant heart rate or motor activity (Almli et al., 2001; DiPietro et al., 2000; Groome et al., 1999), right handedness (Hepper et al., 2005), infant and child development (DiPietro et al., 2002, 2007), and infant temperament (DiPietro et al., 1996; Werner et al., 2007). In developmental psychology research, there is no widely accepted theoretical definition of temperament. Nevertheless, most authors

agree that temperament is a set of genetically influenced traits thought to underpin behavioral individual differences seen after birth (e.g., irritability, emotionality, activity level, or fearfulness) that show some stability over time. According to the existing theories, these predispositions can be seen as more or less influenced by developmental and environmental conditions. Prenatal factors such as maternal stress have been associated with infant temperament (Buitelaar et al., 2003).

In fetal twin studies, there is also some evidence for the emergence of relatively stable inter-individual differences between twins in the first trimester (Degani et al., 2009), the first and second trimester (Piontelli et al., 1999), and in the third trimester of pregnancy (Sherer et al., 1990).

In a prospective study, Sherer et al. (1990) found that the number of FHR accelerations of each twin fetus remained constant throughout the third trimester, suggesting the existence of stable individual differences in FHR by the end of gestation. Degani et al. (2009) have reported an association between the intra-pair difference in total fetal activity observed at 11–14 weeks of pregnancy and the intra-pair difference in infant temperament at 3–6 months of age. According to the authors, maternal reports on infant temperament and the more active twin after birth were correlated with prenatal inter-twin differences in fetal activity.

Inter-pair differences in spontaneous and evoked fetal movements were maintained from 10–11 weeks to 21–22 weeks (Piontelli et al., 1999). In addition, MC twins were found to be more similar than opposite-sex DC twins in both types of fetal movements. Interestingly, this similarity was found to decrease with advancing gestation, suggesting that maternal supply guaranteed by the umbilical cord is not identical for both twins even in normally evolving MC pregnancies. Nevertheless, a sex effect may have accentuated the observed differences in fetal activity levels.

So far, no longitudinal studies have analyzed individual differences in fetal movements throughout gestation. These studies could provide fundamental knowledge about the onset and origins of individual behavioral differences.

Discussion

Thirty years after the publication of the first twin study on fetal behavior reviewed, no definite conclusions can be made about normal fetal movements and heart rate in twin pregnancy. Ultrasound studies comparing fetal activity in twins and singletons are scarce and the results are mixed. Overall, some association or similarity in fetal movement incidences or FHR patterns between both members of twin pairs can be detected. Nevertheless, the temporal occurrence of these patterns is for the most part asynchronous when shorter time intervals were selected. In two of the reviewed studies, it is unclear which criteria were used when the FHR accelerations were called synchronous. A similar problem arises with the determination of synchronous fetal

movements. However, a wide range of estimates is found for simultaneous movement incidence during pregnancy even when a strict definition of synchrony is used (start at the same time). Asynchronous occurrence of rest–activity cycles and different observation periods can partly account for these differences.

From the available research, chorionicity is the only variable that appears to have some influence on fetal behavior. It shows an association with the onset and frequency of inter-twin contacts in early pregnancy, intra-pair differences in fetal movements and FHR during pregnancy. MC twins initiate inter-twin contacts earlier in pregnancy, have more contacts until 13–16 weeks, and demonstrate more similarities than DC twins in fetal movement and FHR patterns. However, differences between MC and DC twin pairs seem to decrease over pregnancy as MC twins become less similar, especially after 30 weeks. In line with these findings, research on the organization of sleep and awake cycles in the most extreme form of monozygosity — the Siamese twin — revealed that a particular behavioral state in one twin member (REM sleep, non-REM sleep, active awake, quiet awake) rarely occurs simultaneously in the co-twin after birth until five months of age (Lenard & Schulte, 1972; Sackett & Korner, 1993). The identical genotype, the shared environment, and the mutually experienced (tactile) stimulation through motor activity do not seem to be able to synchronize the temporal organization of the sleep and awake cycles. Even in the craniopagus, with a common cerebral blood circulation, the different sleep and awake periods occurred almost independent of each other. The existence of a humoral, sleep-inducing factor, which has been suggested based on experiments in animals with coupled circulations, should therefore be doubted (Lenard & Schulte, 1972). Altogether, these findings could indicate that intra-pair similarities in fetal activity expressed in fetal movement incidence and FHR patterns can reflect maturation as previously suggested for fetal behavioral states in the neonatal period (Sackett & Korner, 1993). On the other hand, the intra-pair differences demonstrated on the temporal occurrence of fetal movements and heart rate patterns could be an expression of each twin's individuality.

There is some evidence that the incidence of evoked movements is lower than that of spontaneous movements. This finding corroborates a previous conclusion that fetal movements are, for the most part, independent. It also suggests that not every inter-twin contact provoked by one of the twins is followed by a reaction from the co-twin.

Considering the small number of the reviewed studies that addressed the continuity of individual differences, we can conclude that there is only preliminary evidence for the emergence of relatively stable individual differences in activity and heart rate patterns during pregnancy.

This review highlights the limited number and the high heterogeneity of studies on fetal movements and heart rate patterns in twin pregnancy. The study of fetal behav-

ior in twin and higher order pregnancies is even more time-consuming than in singletons, and also has additional methodological limitations that relate to the monitoring of twins simultaneously, the identification of each twin throughout pregnancy (can be problematic in studies beginning in early pregnancy before sex determination in monoamniotic twins and same-sex twins), the definition of synchrony, conceptual views on twins, and the interpretation of the data. In general, the available research evidence is based on small, selected samples (uncomplicated twin pregnancies), the recording time is (far too) short, and studies were carried out either in the first or the second half of pregnancy with a lack of longitudinal studies. Often, there is no information about the type of placentation or zygosity.

The definitions and procedures used have varied considerably from study to study. Comparison between most of the ultrasound studies describing evoked fetal movements and synchronous/simultaneous fetal movements is hampered by the various concepts and definitions used. Evoked fetal movements include those movements that occur within 1 second of a touch (Arabin et al., 1996) or 4 seconds following an unambiguous stimulus/response pattern in an otherwise immobile fetus (Piontelli et al., 1997). Synchronous/simultaneous fetal movements are thought to be caused either by inter-twin contacts or environmental stimuli. However, the conclusions of some studies of the same group (Sadovsky et al., 1986, 1987; Samueloff et al., 1991) and their a priori assumptions about spontaneous movements and movements induced by maternal and other environmental stimuli have been partly refuted or rendered out of date (Arabin et al., 1995; Visser et al., 1992).

In our opinion, the definitions of the two types of activities — synchronous and evoked fetal movements — and also the different time intervals between the stimulation and the reaction are probably reflecting different aspects of fetal movement patterns of twins. Synchronous activity does not require by definition previous intra-pair stimulation, while the evoked fetal movement include only the activity performed after a stimulus. Some additional problems can also be found when analyzing these definitions. It is not clear if a passive movement following a touch of the co-twin is included in the synchronous and evoked categories. In the ultrasound studies, the qualitative judgment of a body movement can give a definitive answer to the active or passive character of that movement, except probably in the third trimester and under pathological circumstances (large interfetal growth discordancy, whether or not accompanied by oligo- or polyhydramnion). Conversely, with the use of actograph it is not possible to differentiate between active and passive movements because passive movements are simply recorded as fetal activity. The same holds for maternal perception of each twin's fetal movements. Therefore, actograph and maternal perception of fetal movements cannot reliably be used for fetal movement observations in twin studies.

Arabin and van Eyck (2006) have recognized that some reactions could be merely passive movements, non-reactions to the contact of the co-twin that cause displacement of the total body or some body parts due to the intensity of contact. Still it is unclear whether these were included in the 'first reach and touch' pattern proposed by these authors as this movement pattern has been described to disappear at 13 weeks of gestation (Arabin et al., 1996). The absence of a reaction after stimulation, resulting in a passive movement or not, may have a different meaning according to the observed frequency and the stage of pregnancy and thus may be of potential clinical value. There is some evidence that the lack of response of one twin to a contact from the co-twin can be noted from 10–13 weeks onwards (Hata et al., 2011; Piontelli et al., 1997). The underlying causes of this phenomenon are unknown. The absence of response to a contact in late pregnancy in twin pregnancies might be a result of maturation, habituation to repeated contacts, lack of intrauterine space, or a non-responsive sleep state in the co-twin. Decrease in incidence and number of fetal movements has been found in the last trimester of pregnancy both in singletons (ten Hof et al., 2002) and twins (Mulder et al., 2012) and has been interpreted as a developmental phenomenon that could not be simply explained by the appearance of longer episodes of fetal quiescence until term (ten Hof et al., 2002). The use of 4D ultrasound in fetal behavioral studies is another technique that has also several important limitations, especially in the first trimester of pregnancy. Even with the latest 4D ultrasound machines with high frame rates, four movement patterns easily recognized with 2D ultrasound are not detected or documented with 4D ultrasound, namely sideways bending, breathing, hiccups, and facial movements (Kurjak et al., 2006).

Future twin studies could help clarify contradictory results from research with singletons and enhance our knowledge on a number of research topics. In our opinion, these pregnancies are a natural and valuable model to study the potentially adverse maternal psychological (anxiety, stress, depression) and physiological (cortisol) effects on fetal, neonatal, infant, and child behavior and development. Although a direct link has been established between maternal anxiety/stress and fetal behavior in the last trimester of pregnancy in singletons (van den Bergh et al., 2005), there is no such data available from twin pregnancies.

The 'fetal origins hypothesis' of the long-lasting effects of prenatal factors, namely maternal psychological state, on child development has been studied extensively in singletons (Gutteling et al., 2005; Huizink et al., 2004), but not in twins.

There is growing evidence from animal and human studies for a sex-dependent fetal programming by maternal stress (Brunton & Russell, 2010; van den Bergh et al., 2006). Investigations comparing the same-sex with opposite-sex twin pairs could give additional insight into this subject. In

addition, twin studies could provide information regarding the prenatal predictors of maturation and temperament, especially when one of the twins has a recognized malformation or is growth-restricted.

Twin studies on the continuity of movement patterns and heart rate from the prenatal to the postnatal period are also lacking. In singletons, the entire repertoire of fetal movement patterns observed at the third trimester of pregnancy can also be seen after birth (Pechtl, 1984). There is also some evidence for the continuity of heart rate and its variability from prenatal life to postnatal lifespan of 1 year (Almli et al., 2001; Lewis et al., 1970) and 10 years (Thomas et al., 1989).

In addition, appropriately designed twin studies are needed to examine differences in FHR and behavior according to gender as the evidence from singletons is mixed. Although some authors observed gender-specific FHR (Bernardes et al., 2008; Buss et al., 2009) and fetal movements (Almli et al., 2001; Hepper et al., 1997), others have found no gender differences in these prenatal variables (de Vries et al., 1987; DiPietro et al., 2000; Robles de Medina et al., 2003). The same-sex twins were found to have more synchrony in behavior patterns than opposite-sex twins (Gallagher et al., 1992). However, the authors recognize that sex combination could be a confounding variable and chorionicity is probably the explaining cause for this finding. On the other hand, no difference in spontaneous (Arabin et al., 1995) or simultaneous fetal movement (Zimmer et al., 1988) has been found relative to sex combination.

Nevertheless, the considerable interfetal variability of behavior of both singletons and twins has made the identification of abnormal fetus still difficult. These individual differences can be a fundamental topic of research, especially as these have been noticed from early pregnancy onwards and are overall unstable over pregnancy. Methodological aspects, including disregard of the fetal rest–activity and sleep–wake cycles can, at least in part, explain the low intra-individual stability of fetal behavior. Behavioral state-dependent changes in the human fetus have been well documented in the last trimester of pregnancy (Pillai & James, 1990; Visser et al., 1992).

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