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Secular trends in gestational age and birthweight in twins

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BACKGROUND: In recent decades, the overall rate of preterm births has increased. The aim of the present study was to examine whether this trend is also seen for multiple gestations. More specifically, we examined if there has been a decrease in gestational age for live born monozygotic (MZ) and dizygotic (DZ) twins and if there has been a simultaneous change in birthweight. The contributions of fertility treatments and Caesarean sections were taken into consideration. All analyses were carried out in two large European twin cohorts.

METHODS: Cross-sectional study of 6310 live born twin pairs, born between 1964–2007, from the Belgian East Flanders Prospective Twin Survey and 14 712 twin pairs, born between 1990–2006, from the Netherlands Twin Register. Multiple regression analyses were performed with gestational age as outcome variable, and multilevel analysis with birthweight as outcome variable. All analyses were performed with and without adjustment for zygosity, parity, maternal age, mode of conception and delivery and, for the analyses of birthweight, gestational age.

RESULTS: Gestational age decreased in a linear fashion from 1964 to 2007 with a decrease of 0.25 days per year in a similar way for MZ and DZ twins. Changes in birthweight depended on gestational age: up to 32 weeks, birthweight decreased and after 32 weeks birthweight increased. The frequency of infertility treatment and Caesarean sections, primiparity and advanced maternal age increased over the years, but none of these factors influenced the secular trends in gestational age and birthweight.

CONCLUSIONS: The decrease in gestational age and change in birthweight in twins are sources of concern, especially for very preterm twins, for whom birthweight decreased. For twins born after 32 weeks, an increase in birthweight was observed and this is very likely the explanation for the decrease in gestational age.

Key words: twins / gestational age / birthweight / secular trend / time trend

Introduction

One of the causes of adverse neonatal outcome, including neonatal, infant and adolescence morbidity, is preterm birth (Saigal and Doyle, 2008). In recent decades, the overall rate of preterm births has increased, as well as the prevalence of multiple gestations (Tucker and McGuire, 2004; Goldenberg *et al.*, 2008). Multiple gestations carry a substantial risk of preterm delivery and result in 15–20% of all preterm births (Hall, 2003; Tucker and McGuire, 2004; Goldenberg *et al.*, 2008).

In recent decades, the prevalence of preterm birth among singletons and twins has increased in most industrialized countries, while at the same time perinatal mortality has decreased, mainly because of medically indicated preterm birth (Joseph *et al.*, 1998, 2001; Kogan *et al.*, 2000; Ananth *et al.*, 2004a, b; Ananth *et al.*, 2005; Kramer *et al.*, 2005; Goldenberg *et al.*, 2008; Keirse *et al.*, 2009; Hartley and Hitti, 2010). Whereas it is apparent that gestational age in recent decades for twins and singletons has become shorter, a similar trend is less obvious for birthweight. In general, birthweight of singletons has increased, especially in children born at term (Power, 1994; Kramer *et al.*, 2002; Wen *et al.*, 2003; Davidson *et al.*, 2007), suggesting a trend of improved ‘fetal growth’. This trend may be explained by increases in maternal anthropometry, reduced cigarette smoking and changes in sociodemographic factors (Kramer *et al.*, 2002). In contrast, for preterm births birthweight decreased (Power, 1994; Wen *et al.*, 2003), (Branum and Schoendorf, 2002; Kramer *et al.*, 2005). Also for twins, the prevalence of term small for gestational age (SGA) twins has decreased (Kogan *et al.*, 2000; Joseph *et al*., 2001), whereas the prevalence of (very) low birthweight [(V)LBW] (Ananth *et al*., 2004a, b) and preterm SGA has increased (Kogan *et al*., 2000).This may be the consequence of improved perinatal care, where twins at risk are detected earlier and where obstetric interventions lead to preterm birth.

Investigations of secular trends over a long period of time (e.g. 40 years) can be biased by changes in obstetrical and neonatal care. For example, there has been an increase in the use of ultrasound to measure gestational age. Since both pre- and post-term birth rates are overestimated if gestational age is based on the last menstrual period only (Joseph *et al*., 2007), using ultrasound results in both shorter and longer gestational ages. Additional changes over time include the introduction of artificial reproductive technology (ART) around 1980; a change in obstetrical policies due to technological advances, e.g. antenatal corticosteroids and intensive care for premature neonates; and changes in socio-demographic variables (Kramer *et al*., 1998) including maternal age. At present, almost half of all twin pairs in Belgium and the Netherlands are born after infertility treatment (Loos *et al*., 1998; Derom *et al*., 2006; Gielen, 2007) and Dutch mothers are among the oldest in the world (Statistics Netherlands, www.cbs.nl).

To date, no study addressed the question whether there are differences in secular trend between dizygotic (DZ) twins and monozygotic (MZ) twins [monochorionic (MC) or dichorionic (DC)], or differences between naturally and medically conceived twins. These are relevant questions as MZ twins have lower birthweights than DZ twins, MC twins have lower weights than DC twins (Gielen *et al*., 2007, 2008) and twins resulting from ART (mostly DZ) have an increased risk of preterm birth (Verstraelen *et al*., 2005). The mother of twins born after ART is more often a primipara of advanced age (Delbaere *et al*., 2008).

The aims of the present study were to determine whether there has been a decrease in gestational age for twins as a function of zygosity and a simultaneous change in birthweight, and to evaluate the contribution of chorionicity, fertility treatment and Caesarean sections in two large European twin cohorts: first, a Belgian population-based cohort in which data collection took place from 1964 to 2007 and second, a Dutch population-based volunteer cohort in which data were available for birth cohorts 1987–2006.

Materials and Methods

East Flanders Prospective Twin Survey

The East Flanders Prospective Twin Survey (EFPTS), which started in 1964, is a population-based register of multiple births in the province of East Flanders (Belgium). The twins (and higher order births) are ascertained at birth. Basic perinatal data are recorded and chorion type and zygosity are determined. The study sample consisted of 7500 twin pairs, who met the World Health Organization criteria for live born infants (birthweight $\geq$500 g or gestational age $\geq$22 weeks, if birthweight unknown), selected from the EFPTS in Belgium (Loos *et al*., 2005) between July 1964 and the end of December 2007.

Data recorded by the obstetrician at birth included gestational age, birthweight, maternal age, parity, mode of delivery and mode of conception, i.e. naturally conceived versus ART (ovulation stimulation and *in vitro* fertilization IVF with or without intracytoplasmic sperm injection (ICSI) and live born (including early neonatal death) versus stillborn. Gestational age was based on the last menstruation or on a first trimester ultrasound investigation (only available during the second half of this study), and was calculated as the number of completed weeks of pregnancy. Zygosity was determined by sequential analysis based on sex, fetal membranes (chorionicity), umbilical cord blood groups, placental alkaline phosphatase, and, since 1982, DNA fingerprints (Vlietinck, 1986). After DNA-fingerprinting, a zygosity probability of 0.999 is reached. MC twins and same-sexed DC twins with the same markers were classified as MZ (Loos *et al*., 1998).

Information on ethnicity was obtained from the Study Center for Perinatal Epidemiology (SPE), a comprehensive clinical birth database of Flanders (Cammu *et al*., 2006) (personal communication, 2006). The prevalence of Caucasians in the province of East Flanders is over 99%, even with an increased prevalence of people of foreign descent since 1964. In the two most recent years, 84.5% of the population was of Belgian origin, 5% of Turkish and 2.3% of Moroccan origin. The remaining 8.2% was of other origin (mainly Caucasians of Dutch, French and German origin) or unknown origin.

Twin pairs of whom one or both children were stillborn (252 pairs) were excluded. Twin pairs with missing or inconsistent data [gestational age (593 pairs), parity (54 pairs), mode of conception (69 pairs), maternal age (59 pairs), zygosity (140 pairs) and birthweight (27 pairs)] were also excluded. In total, data from 12 620 twins (6310 pairs) were analysed: 2048 MZ twin and 4262 DZ twin pairs, including 2092 opposite-sex twin pairs. The percentage of boys was 50.6%.

Netherlands Twin Register

Since 1987 the Netherlands Twin Register (NTR), a population-based volunteer register, recruits families with twins a few months after birth and registers around 40% (ranging from 38 to 53%) of all multiple births in the Netherlands. The NTR is maintained by the Department of Biological Psychology at the VU University, Amsterdam (Boomsma *et al*., 2002, 2006; Bartels *et al*., 2007).

Shortly after registration of their newborn twins, mothers receive a survey in which they are asked to report on mode of delivery, age at birth, gestational age, birthweight, birth order, sex of twins and mode of conception (Survey 1). Information on birth delivery mode was not available for twins born before 1990. Information on parity was obtained by a two-page survey about familial twinning that was sent to all mothers of twins who were registered with the NTR in 2005 (C.Hoekstra *et al*., submitted) and by a survey sent to the mother when the twins were 5 years old. For 16 700 families, Survey 1 and data on parity were available. Families were excluded when data on zygosity (209 pairs), mode of conception (635 pairs), delivery (959 pairs), gestational age (99 pairs) and maternal age (86 pairs) were missing or inconsistent.

Information on ethnicity was based on the country of birth of the mother: 96.6% were born in the Netherlands, 0.5% in the Antilles and Surinam, 0.1% in Morocco, 0.2% in Turkey, 1.8% were born in a western non-Dutch country and 0.7% in a non-western country.

For same-sex twin pairs, zygosity was based on DNA polymorphisms obtained in previous NTR studies ($n = 767$ pairs) or from survey questions on physical resemblance and frequency of confusion of the twins by family and strangers (6745 pairs). The correspondence between questionnaire and DNA zygosity is 93% (Rietveld *et al*., 2000). When DNA and previous survey data were not available, zygosity was based on the mother report in the survey shortly after birth ($n = 2318$). The final sample included 29 424 twins (4643 MZ twin pairs and 10 069 DZ twin pairs; including 4882 opposite-sex twin pairs) from birth cohorts 1990–2006. The percentage of boys was 49.8%.

Definitions and reference data sets

LBW was defined as birthweight less than 2500 g; VLBW as <1500 g. Preterm birth was defined as born before 37 weeks, and very preterm birth as before 32 weeks. For the EFPTS, this was up to 36 weeks plus 6 days and up to 31 weeks plus 6 days and for the NTR up to 36 $\frac{1}{2}$ weeks (=36 weeks plus 4 days) and up to 31 $\frac{1}{2}$ weeks (=31 weeks plus 4 days). Early neonatal death was defined as death within the first week of life. SGA was defined as less than the 10th centile, with singletons of the local population as reference. For the EFPTS data of the SPE (Cammu *et al.*, 2006) were used as reference (personal communication, 1999). For the NTR data of the Netherlands Perinatal Registry (NPR) (Stichting Perinatale Registratie Nederland (NPR), 2006), which registers all births in the Netherlands, (personal communication, 2008) were used. Birthweights of the NTR were also compared with the birthweights of live born twins between 2000 and 2006 ($n = 48.462$) from the NPR. However, zygosity could not be obtained from this database.

Statistical analyses

Comparisons of the characteristics between the MZ and DZ twins and between the EFPTS and NTR cohorts were performed by χ^2 tests for comparisons of frequencies, while for comparing means an *F*-test followed by the appropriate *t*-test was used.

The use of ultrasound to determine gestational age was not available until 1985, while the introduction of ART started in the early 1980s. The analyses to study a secular trend in gestational age and birthweight were performed on three data sets of live born twins (including early neonatal death) from the EFPTS. The first data set included all twins between 1964 and 2007, the second only naturally conceived twins in 1964–2007 and the third all twins in 1984–2007. Data from twins from the NTR (born between 1990 and 2006) were analysed twice: once for all twins and once for only the naturally conceived live born twins.

To study a secular trend in gestational age (a pair-level trait), univariate and multiple linear regression analysis were performed with gestational age as outcome variable and with zygosity, parity (primiparity versus multiparity), maternal age, mode of delivery (vaginal versus Caesarean section) and mode of conception (naturally conceived versus ART) as predictor variables. In addition for the EFPTS, chorionicity was incorporated in the model and for the NTR, ethnicity. To evaluate a time trend in birthweight a multilevel analysis (proc mixed) was performed taking into account clustering of the twin pairs by adding a random effect to the model. The variance-covariance structure for birthweight was allowed to differ between MZ and DZ pairs. These analyses were performed with and without adjustment for the predictor variables and with gestational age as an additional covariate.

Next, quadratic terms were included in the models. Interactions between the covariates and year of birth were tested. In the NTR cohort, ART interacted with year of birth ($P = 0.025$), but not when analyzing MZ and DZ data separately. Therefore, we have not included this interaction in the model.

The analyses were conducted with the SAS version 9.1 software package (SAS Institute Inc., Cary, NC, USA). All reported *P*-values are two-sided and were considered statistically significant when $P \leq 0.05$.

Results

Characteristics

Gestational ages were shorter in the later cohorts and were shorter for the Belgian EFPTS twins than for the Dutch NTR twins. In addition, MZ twins had shorter gestational ages than DZ twins. The prevalence of preterm deliveries increased in the most recent (1985–2007) EFPTS cohort compared with the early (1964–1984) EFPTS cohort (Table I).

Birthweights were lower in the later cohorts and were lower for the Belgian EFPTS twins than for the Dutch NTR twins. The prevalence of (V)LBW twins was higher in the most recent (1985–2007) EFPTS cohort than in the early (1964–1984) EFPTS cohort. In addition, MZ twins had lower birthweights than DZ twins. In the 1985–2007, EFPTS cohort SGA was less frequent after 32 weeks but more frequent before 32 weeks of gestation than in the 1964–1984 EFPTS cohort (Table I).

The proportion of MC twins was the same for both EFPTS cohorts. Between 1964 and 1984, the proportion of MC twins for (very) preterm birth, (V)LBW and SGA was higher than the expected proportion of 0.67. Between 1985 and 2007, the proportion of MC did not differ substantially from 0.65, except for ART and SGA before 37 weeks. The prevalence of early neonatal death was lower in the most recent cohort compared with the early cohort, especially if born very preterm. But for the MC twins, neonatal death was higher than the expected proportions of 0.67–0.65 resp. in both EFPTS cohorts (Table I).

In the most recent cohorts (EFPTS cohort of 1985–2007 and the NTR cohort), the prevalence of Caesarean sections was higher and twins were more often born after ART than in the early cohort (EFPTS cohort of 1964–1984). Both MZ and DZ twins had older mothers in the later cohorts. Maternal age is higher in the Netherlands than in Belgium (Table I).

Unadjusted mean gestational age and birthweight

The unadjusted mean gestational age decreased from 1964 to 2007 and was higher for Dutch twins than for Belgian twins. It seemed that the mean unadjusted birthweight also decreased from 1964 to 2007, but this decrease was far less obvious than the decrease in gestational age. Mean birthweights of the Dutch twins were higher than those of the Belgian twins. For both cohorts, gestation lasted longer and birthweights were higher for DZ twins than for MZ twins.

Regression analysis: gestational age

There was a linear decrease of gestational age. All predictor variables contributed to the models with gestational age as outcome variable, except for chorionicity, maternal age for the EFPTS cohort of spontaneous pregnancies and mode of delivery for the NTR cohorts (Supplementary Table SA1). As expected, DZ twins had longer gestational ages than MZ twins, but the secular time trend was not influenced by zygosity, nor by ART, parity or maternal age. In Belgium, Caesarean sections shortened gestational age over time, but not after 1985.

For the estimation of the adjusted mean gestational age of each year, the complete time period from 1964 to 2007 was analysed. The decrease of the adjusted mean gestational ages for four different cohorts is shown in Fig. 1. For Belgian twins, the adjusted mean gestational age of the naturally conceived DZ twins decreased less than the gestational age of all DZ twins, indicating that ART pregnancies were shorter and that the number of ART pregnancies increased over time, resulting in a lower mean adjusted gestational age. For Dutch twins, the same secular trend is observed, i.e. DZ twins born after ART had shorter gestational ages than twins born after spontaneous pregnancies (Fig. 1).

Table I Characteristics of live born twins of the Belgian EFPTS (*n* = 12 620; 6310 pairs) and the Dutch NTR (*n* = 29 424; 14 712 pairs).

	EFPTS, Belgium (1964–1984)			EFPTS, Belgium (1985–2007)			NTR, the Netherlands (1990–2006)	
	MZ (*n* = 1548) (pairs = 774)	MC (Proportion = 0.67)	DZ (*n* = 1982) (pairs = 991)	MZ (*n* = 2548) (pairs = 1274)	MC (Proportion = 0.65)	DZ (*n* = 6542) (pairs = 3271)	MZ (*n* = 9286) (pairs = 4643)	DZ (*n* = 20 138) (pairs = 10 069)
Mean (SD)	Pairs							
Gestational age	36.7 (3.0)	36.6 (3.0)	37.0 (2.9)	35.8 (2.8)***	35.8 (2.8)	36.0 (2.5)***	36.3 (2.6)$^{\$\$\$}$	36.7 (2.4)$^{\$\$\$}$
Maternal age	26.4 (4.9)	26.5 (4.8)	27.7 (4.2)	28.4 (4.6)***	28.4 (4.4)	29.7 (4.4)***	30.7 (3.9)$^{\$\$\$}$	31.7 (3.8)$^{\$\$\$}$
Individuals								
Birthweight	2423 (546)	2381 (533)	2549 (544)	2377 (556)*	2372 (530)	2453 (530)***	2423 (542)$^{\$\$}$	2533 (550)$^{\$\$\$}$
Frequency (%)	Pairs	Proportion			Proportion			
Preterm	299 (39%)	0.69	343 (35%)	677 (53%)***	0.65	1714 (52%)***	1807 (39%)$^{\$\$\$}$	3355 (33%)$^{\$\$\$}$
Very preterm	45 (5.8%)	0.71	51 (5.2%)	96 (7.5%)	0.66	168 (5.1%)	275 (5.9%)$^{\$}$	440 (4.4%)
C-section	58 (7.5%)	0.57	95 (9.6%)	382 (30%)***	0.64	1251 (38%)***	1231 (27%)$^{\$}$	3116 (35%)$^{\$\$\$}$
ART	8 (1.0%)	0.63	63 (6.4%)	77 (6.0%)***	0.77	1715 (52%)***	199 (4.3%)$^{\$}$	3471 (35%)$^{\$\$\$}$
Primiparity	373 (48%)	0.65	397 (40%)	599 (47%)	0.64	1745 (53%)***	2259 (49%)	5234 (52%)
Individuals								
LBW	789 (51%)	0.70	832 (42%)	1414 (55%)**	0.65‡	3268 (50%)***	4242 (46%)$^{\$\$\$}$	7903 (40%)$^{\$\$\$}$
VLBW	90 (5.8%)	0.71	66 (3.3%)	178 (7.0%)	0.65	323 (4.9)**	546 (5.9%)$^{\$\$\$}$	826 (4.2%)$^{\$\$}$
SGA	662 (43%)	0.70	721 (36%)	780 (31%)***	0.65‡	1791 (27%)***	2417 (26%)$^{\$\$\$}$	4936 (25%)$^{\$\$\$}$
≥37 weeks	448 (61%)	0.69	524 (51%)	327 (51%)**	0.64	804 (48%)	1902 (38%)$^{\$\$\$}$	4047 (33%)$^{\$\$\$}$
≥32–37 weeks	105 (21%)	0.78	103 (18%)	219 (19%)	0.68	495 (16%)	501 (14%)$^{\$\$}$	866 (13%)$^{\$\$\$}$
<32 weeks	3 (3.3%)	0.33	4 (3.9%)	10 (5.2%)	0.70	25 (7.4%)	14 (2.6%)$^{\$}$	23 (2.6%)$^{\$\$\$}$
Neonatal death	52 (3.4%)	0.79	64 (3.2%)	50 (2.0%)**	0.70	60 (0.9%)***	NA	NA
≥37 weeks	0		5 (0.5%)	2 (0.3%)	1.00	2 (0.1%)	NA	NA
≥32–37 weeks	14 (2.8%)	0.79	13 (2.2%)	9 (0.8%)**	0.78	13 (0.4%)***	NA	NA
<32 weeks	37 (41%)	0.78	46 (45%)	37 (19%)***	0.70	42 (12.5%)***	NA	NA

MZ, monozygotic; DZ, dizygotic; MC, monochorionic; LBW, low birthweight; VLBW, very low birthweight; SGA, small for gestational age; C-section, Caesarean section; ART, artificially reproductive technique; NA, not available [ovulation stimulation and *in vitro* fertilization IVF with or without intracytoplasmic sperm injection (ICSI)].
Comparisons of two cohorts for MZ and DZ separately.
(1) 1964–1984 to 1985–2007 of the EFPTS: **P* = 0.01; ***P* ≤ 0.005; ****P* < 0.0001.
(2) 1985–2007 of the EFPTS to the NTR: $^{\$}P \leq 0.05$; $^{\$\$}P \leq 0.005$; $^{\$\$\$}P < 0.0001$.
Comparison of proportion MC 1964–1984 to proportion MC 1985–2007: $^{\ddagger}P < 0.05$.

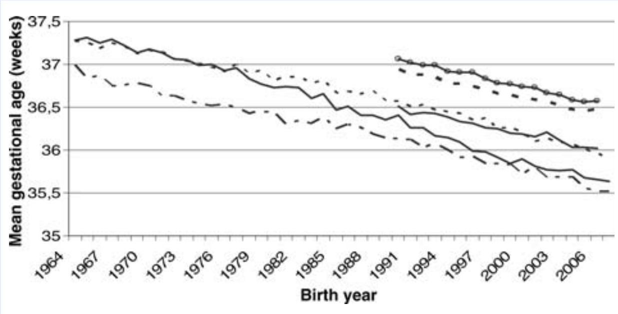

Figure 1 Mean gestational ages for all live born monozygotic (MZ) and dizygotic (DZ) twins and for naturally conceived MZ and DZ twin pregnancies. Legend: adjusted for year of birth, mode of conception, mode of delivery, zygosity, parity and maternal age. ———, All DZ twins of the Belgian EFPTS. – – – –, MZ twins of the Belgian EFPTS. ………… DZ naturally conceived twins of the Belgian EFPTS. ▪▪▪▪▪▪ All DZ twins of the Dutch NTR. ▬▬▬ MZ twins of the Dutch NTR. —○— DZ naturally conceived twins of the Dutch NTR.

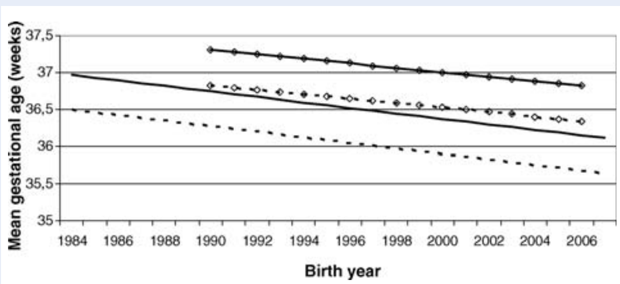

Figure 2 Model for gestational age from 1984 to 2007 of all live born monozygotic (MZ) and dizygotic (DZ) twins. Legend: presented curves are for a naturally conceived pregnancy, vaginal delivery, multiparity and mean maternal age (29 years). ——— DZ twins of the Belgian EFPTS. – – – – MZ twins of the Belgian EFPTS. —◇— DZ twins of the Dutch NTR. – ◇ – – MZ twins of the Dutch NTR.

Because the use of ultrasound and ART was not available during the first half of this study, we present the curves of the models for gestational age and birthweight for the EFPTS cohort of 1985–2007 and the NTR cohort. Figure 2 shows the decrease in gestational age for the EFPTS cohort of 1984–2007 and the NTR cohort for naturally conceived pregnancies of a multipara of 29 years and vaginal delivery. The decrease was the same for both cohorts: around 5 days in 20 years [EFPTS: 0.037 weeks (0.26 days) per year versus NTR: 0.030 weeks (0.21 days) per year], as was the difference between MZ and DZ twins (EFPTS: 3.29 days versus NTR: 3.37 days). Overall, gestation for the NTR lasted over 4 days longer. For the NTR, ethnicity did not additionally influence the trend in gestational age ($\beta = 0.02$ $P = 0.70$).

Multilevel regression analysis: birthweight

Although birthweight decreased in a linear trend from 1964 to 2007, this decrease was not present in the EFPTS cohort of 1985–2007 if adjusted for all covariates except gestational age (Supplementary Table SB1). A decrease in gestational age from 1985 to 2007 was not accompanied with a decrease in birthweight, indicating a general trend towards higher birthweights for a given gestational age. Whereas from 1964 to 2007 MC twin pairs had lower birthweights than DC twin pairs, the difference was not apparent in the cohort from 1985 to 2007. For the Dutch pregnancies, birthweight decreased over time (Supplementary Table SB1).

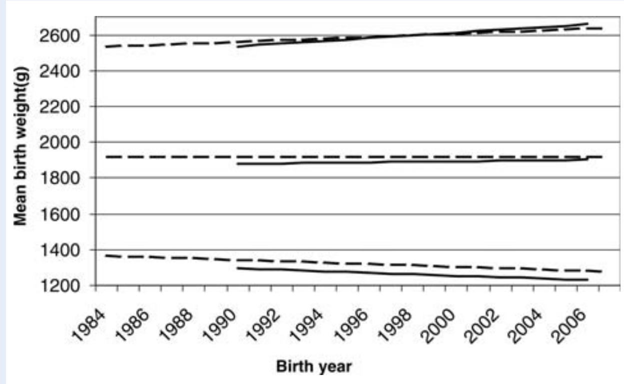

Figure 3 Model for birthweight controlled for gestational age from 1984 to 2007 of all live born dizygotic (DZ) twins. Legend: the presented curves are for a naturally conceived pregnancy, multiparity and mean maternal age (29 years). The dashed curve represents the Belgian EFPTS, the straight curve the Dutch NTR. The two upper curves are for a DZ twin born at 36.5 weeks of gestation, the middle two curves are for a DZ twin born at 32 weeks of gestation, and the lower two curves are for a DZ twin born at 28 weeks of gestation.

Figure 3 (for estimates see additional Table SB2) shows the change in birthweight for the EFPTS cohort of 1984–2007 and the NTR cohort after controlling for gestational age in addition to all other covariates. For both cohorts, birthweight decreased up to around 32 weeks of gestation (EFPTS: 32 weeks versus NTR: 31 weeks), remained the same at 32 weeks (EFPTS) or at 31 weeks (NTR), and increased after those weeks of gestation. The difference between MZ and DZ twins was about the same for both cohorts: 58.9 g for the NTR and 50.1 g for the EFPTS (curves of MZ twins are not shown). The curves of birthweights of both cohorts were close to each other, although NTR twins born in 2006 were 51.3 g lighter at 28 weeks of gestation than the EFPTS twins and, at 32 weeks of gestation 14.5 g and at 36.5 weeks of gestation 27.0 g heavier. With an average increase in birthweight of 150 g per week (Gielen *et al.*, 2008), 51.3 g corresponds approximately to 2.4 days of gestation, indicating that the very premature Dutch twins are heavier than the Belgian twins for a given gestational age. Again, for the EFPTS, the influence of chorionicity disappeared in the cohort from 1985 to 2007.

Discussion

Over the past decades, there has been a decrease in gestational age in twins, which is of the same magnitude for MZ and DZ twins. A very similar trend was seen in two large European twin cohorts from

Belgium and the Netherlands. With a decrease of ~0.25 days per year, this has led to a decrease of 5 days over the past 20 years. For birthweight, we observed a change that depended on gestational age. Up to 32 weeks of gestation birthweight decreased over time, if controlled for gestational age, and increased after 32 weeks. This is consistent with prior studies (Kogan *et al.*, 2000; Joseph *et al.*, 2001; Ananth *et al.*, 2004a, b).

Twins born after ART and/or Caesarean sections had shorter gestational ages, but an increase in ART and Caesarean sections could not completely explain the secular trend in gestational age. Importantly, we have no indications that this trend has reached the end. While the end-point of the previous reported studies was before 2000 (Kogan *et al.*, 2000; Joseph *et al.*, 2001; Ananth *et al.*, 2004a, b) 2002 (Keirse *et al.*, 2009) or 2005 (Hartley and Hitti, 2010), the end-point of this study was 2007, indicating that this secular trend is still ongoing.

Two possible explanations for the secular trend in gestational age can be found in the change in birthweight, which is different for twins born after 32 weeks and very preterm births. First, the adjusted birthweight increased for twins born after 32 weeks of gestation, which is consistent with other studies that showed a decrease of the prevalence of term SGA twins (Kogan *et al.*, 2000; Joseph *et al.*, 2001). The decrease in gestational age is probably due to the fact that twins are getting bigger, at least after 31 weeks of gestation. This causes uterine overdistension, one of the causes of preterm delivery (Romero *et al.*, 2006), which may lead to parturition and therefore shorter gestational ages. For term births in singletons, reduced cigarette smoking, higher educational levels (Kramer *et al.*, 1998; Kim *et al.*, 2007), other changes in socio-demographic factors (Kramer *et al.*, 2002) and an increase in maternal anthropometry (Kramer *et al.*, 2002; Kim *et al.*, 2007) seem to explain the trend for birthweight. One would expect that these factors also explain part of the trend observed for twins. For a part of the NTR cohort information on educational level, smoking behaviour during pregnancy and BMI were available. Of these three factors only educational level was significantly associated with gestational age, higher educational levels being associated with longer gestational ages. We found no evidence for an interaction between any of these factors and year of birth. Thus, it seems that these factors do not explain the secular trend of gestational age.

Second, birthweight decreased for twins born very prematurely. Earlier studies also showed that the prevalence of (V)LBW (Ananth *et al.*, 2004a, b) and preterm SGA increased (Kogan *et al.*, 2000). The decrease in birthweight could be due to higher survival rates of SGA twins born very preterm, because of advanced obstetrical and neonatal procedures. Therefore, clinicians will more often be confronted with survivors of very preterm birth. This will also lead to higher birthweights after 32 weeks, when the fetuses at risk have already been born. Although the covariates in our study did not explain the trend in gestational age, additional observations may demonstrate that advanced obstetrical and neonatal procedures have led to lower mortality rates. First, if the NTR twins had been compared with the standards (with references of the 1960s) of Kloosterman (1970), the prevalence of SGA <32 weeks would have been higher (11% for MZ and 8.5% for DZ twins), whereas the prevalence of SGA for twins born after 32 weeks would have been the same. Second, for the EFPTS cohort, the prevalence of neonatal death and of stillborn twins (data not shown) decreased over the years with a simultaneous decrease of gestational age. Birthweight (controlled for gestational age) also decreased for the stillborn twins (data not shown). For the live born twins of the EFPTS cohort 1985–2007, gestational age and birthweight decreased by the same amount for surviving twins and twins who died neonatally. Meanwhile, the prevalence of Caesarean sections was higher in survivors than in twins with early neonatal death (Gielen *et al.*, 2008).

We have limited data on late neonatal, infant or adolescent morbidity. However, it is known that the prevalence of cerebral palsy is inversely related to gestational age (Saigal and Doyle, 2008). A reduction in neonatal mortality will therefore result in an increase in absolute numbers of cases of cerebral palsy as a function of an increase in neonatal survival (Saigal and Doyle, 2008). Besides the fact that severe neurological and developmental disability is common among extremely premature survivors (Wood *et al.*, 2000), the risks of medical and social disabilities in adulthood increased with decreasing gestational age (Moster *et al.*, 2008). Twinning is not a rare event, causing considerable health care problems in Western societies. Tyson *et al.* (2008) proposed that the likelihood of a favourable outcome can be better estimated by consideration of four additional factors to gestational age: the effect of female sex, exposure to antenatal corticosteroids, being a singleton (compared with being a multiple), and 110 g higher birthweight; they were each of the same magnitude as 1 week of gestational age.

Differences in birthweight and gestational age between MZ and DZ twins have remained the same over the past decades and chorionicity did not seem to influence the secular trend in the decrease of gestational age. One might have expected that these differences between MZ and DZ twins would have become smaller over the past decades, because of advanced obstetrical interventions. Two-thirds of the MZ twins are MC and are therefore at risk of twin to twin transfusion syndrome (TTTS). Because only live born twin pairs were analysed in this study, we might have missed severe TTTS twin pairs, of whom one or two twins have died. Nonetheless, the frequency of neonatal death among MC twins was higher then expected, indicating that, despite advanced obstetrical interventions, mainly MC twins are still at risk of neonatal death. Furthermore, gestational age of stillborn twins also decreased from 1984 to 2007 with a higher prevalence of MZ twins (2.53 versus 0.63% for DZ twins), and more especially MC twins (EFPTS, data not shown).

It is remarkable that the decrease in gestational age, as well as the difference between MZ and DZ twins, was of the same magnitude in Belgium and the Netherlands, despite the differences in data collection of the two registers. Whereas the EFPTS ascertains all twins at birth, the NTR registers around 40% of all multiple births, and recruits families a few months after birth. Twins with early neonatal death or twin pairs of whom one or both twins suffer from severe morbidity (lower birthweights and shorter gestational ages) might therefore be underrepresented in the NTR. To test this, gestational ages and birthweights were compared with a reference data set of all Dutch live born twins (NPR). Gestational age and birthweights of the NTR were higher then those of the NPR, while for the EFPTS gestational age was the same and birthweights were lower (NPR: 35.9 (3.0) weeks and 2459 (615) g; NTR: 36.5 (2.4) weeks ($P < 0.01$) and 2498 (550) g ($P < 0.01$); EFPTS: 35.9 (2.6) weeks and 2432 (539) g ($P = 0.004$)). This confirms the presence of selection bias of the NTR, but it is also consistent with the fact that Dutch people are taller than Belgian people, which could also explain the higher birthweights for Dutch twins.

One limitation of this study is the lack of individual data and consistency over time on the method of estimating gestational age. In the Belgian cohort, last menstrual period was used during the first half of the study period, while ultrasound or last menstrual period was used during the later period. In Belgium, first trimester ultrasound dating was routine already in the 1980s. In the Dutch cohort, gestational age was based on maternal recall after delivery. In the Netherlands, the estimation of gestational age will have changed over time. Obstetrical care is initially provided by midwives and general practitioners. When risk factors are recognized, such as a twin pregnancy, pregnant women are referred to an obstetrician (Amelink-Verburg *et al.*, 2008). Although already used in some primary care centres, first trimester ultrasound became routine in the Netherlands in January 2004 when a reimbursement system was introduced.

The different methods of estimating gestational age may explain the differences in gestational age between the Belgium and Dutch cohort's. However, although the use of ultrasound will result in a more exact estimation of gestational age, the ultrasound dating will give a reduction of pre and post-term birth (Joseph *et al.*, 2007). In this respect, we expect no systematic bias because of the use of ultrasound. On the other hand, ultrasound dating will lead to better monitoring and identifying twins at risk and therefore to medically induced labour with shorter gestational ages. Although at present no clear benefit in terms of perinatal mortality can be discerned to result from the use of ultrasound, one could speculate that routine application of ultrasound at the end of the first trimester of pregnancy might likewise have effected in a substantial reduction of perinatal mortality (De Reu *et al.*, 2000). In addition, Hartley and Hitti reported an increase in the need for respiratory support in twins along with an increase of preterm birth and a decrease of neonatal mortality (Hartley and Hitti, 2010). This could be another explanation for the decline in gestational age. In this respect, one would expect a larger reduction of gestational age in the Dutch cohort than the Belgium cohort. However, we found no evidence for a different slope of gestational age between the Dutch and Belgium cohort.

In summary, fetal surveillance has improved, leading to a better identification of fetuses at risk. Caesarean sections will be performed in these cases of high risk, and obviously in cases with shorter gestational ages and lower birthweights (IUGR). This effect is not present after 32 weeks, when the fetuses at risk are already born. If the course of pregnancy is undisturbed up to 37 weeks, this will lead to well grown twins, comparable with the data in singletons. Both higher and lower birthweights are a source of concern and the clinicians have to deal with the increasing morbidity of survivors of (very) preterm birth. It is also another reason to aim at a reduction in the incidence of twin pregnancies, certainly in patients undergoing ART. In the Netherlands, single embryo transfer (SET) started in 2005 and in Belgium a reimbursement system for SET has been set up since 2003 (Gerris, 2005), resulting in a decrease of the frequency of twin pregnancies.

Authors' roles

All authors of this research paper have directly participated in the planning and execution, have approved the final version submitted and have no conflict of interest in connection with this paper. M.G. had full access to the data of the EFPTS, C.E.M.B. to the data of the NTR. They take responsibility for accurate data analysis. C.D. and R.V. take responsibility for the integrity of the data collection of the EFPTS. D.I.B. takes responsibility for the integrity of the data collection of the NTR. Study concept and design: D.I.B., C.D. and M.P.A.Z. Analysis interpretation of data: M.G., C.E.M.B., C.D., R.V., J.G.N., M.P.A.Z. and D.I.B. Drafting the manuscript: M.G. and C.E.M.B. Critical revision of the manuscript for important intellectual content: M.G., C.E.M.B., C.D., R.V., J.G.N., M.P.A.Z. and D.I.B. Statistical analysis: M.G. and C.E.M.B. Study supervision: D.I.B., C.D. and M.P.A.Z.

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Supplementary data

Supplementary data are available at http://humrep.oxfordjournals.org/.

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