Body composition, smoking, and spontaneous dizygotic twinning

Chantal Hoekstra, M.Sc.,[a] *Gonneke Willemsen, Ph.D.,*[a] *C. E. M. Toos van Beijsterveldt, Ph.D.,*[a] *Cornelius B. Lambalk, M.D.,*[c] *Grant W. Montgomery, Ph.D.,*[b] *and Dorret I. Boomsma, Ph.D.*[a]

[a] Department of Biological Psychology, VU University Amsterdam, Amsterdam, The Netherlands; [b] Molecular Epidemiology Laboratory, Queensland Institute of Medical Research, Brisbane, Australia; and [c] Division of Reproductive Medicine, VU Medical Centre, Amsterdam, The Netherlands

Objective: To examine differences in body composition and smoking between mothers of spontaneous monozygotic and dizygotic twins, while taking into account maternal age, gravidity, and educational attainment.
Design: Retrospective cohort study.
Setting: The Netherlands Twin Register.
Patient(s): Mothers of twins (n = 19,357) registered with the Netherlands Twin Register. Data were selected from mothers of spontaneous monozygotic twins (MZ; n = 5663) and mothers of spontaneous dizygotic twins (DZ; n = 8515).
Intervention(s): None.
Main Outcome Measure(s): The odds of having spontaneous DZ twins versus spontaneous MZ twins as a function of height, body mass index (BMI), and smoking before pregnancy, after accounting for age, gravidity, and educational attainment.
Result(s): Compared with spontaneous MZ twinning, spontaneous DZ twinning is significantly associated with increasing height (odds ratio, 1.6; 95% confidence interval [CI], 1.5–1.8 for the tallest versus the shortest height quartile), an increased BMI (odds ratio, 1.3; 95% CI, 1.1–1.4 for overweight vs. normal weight), and smoking before the twin pregnancy (odds ratio, 1.4; 95% CI, 1.3–1.5 for smoker vs. nonsmoker). Maternal age and gravidity, but not educational attainment, had to be included in the model.
Conclusion(s): Spontaneous dizygotic twinning is associated with body composition and smoking. (Fertil Steril® 2010;93:885–93. ©2010 by American Society for Reproductive Medicine.)

Key Words: Body composition, smoking, educational attainment, maternal age, gravidity, multiple pregnancies

Twinning rates vary across the world, from six per 1000 births in Asia to 40 per 1000 births in Nigeria (1–4). Twinning rates also vary over time. For example, over the last 30 years Dutch twinning rates increased from 10 per 1000 pregnancies to 18 per 1000 pregnancies (4, 5). The regional and time-dependent differences are mainly due to differences in dizygotic (DZ) twinning rates (6). Monozygotic (MZ) twinning occurs around the world at a constant rate of around three to four per 1000 births and presents a random process that may happen to any woman having children (2, 4, 6–9). In contrast, DZ twinning is influenced by genetic, maternal, and environmental factors. Family history and increased parity or gravidity are known to increase the risk of spontaneous DZ twinning (1, 2, 4, 7, 10, 11). The increase in DZ twin births seen in the Netherlands and other Western countries in the last decades can be attributed to increases in the number of fertility treatments but also to increases in maternal age (12–14).

Established risk factors do not explain all variation in DZ twinning, and additional factors are likely to be involved. These may include body composition and smoking. In a study by Basso et al. (15), mothers of twins (MZ and DZ) were taller and had a higher body mass index (BMI) than the mothers of singletons. Information on the zygosity of the same-sex twin pairs was not available, but the association became more pronounced when singleton mothers were compared with opposite-sex twin mothers (i.e., DZ twin mothers). In accordance with these findings, Reddy et al. (16) compared MZ and DZ twin mothers to singleton mothers

Received January 16, 2008; revised September 27, 2008; accepted October 11, 2008; published online December 4, 2008.
C.H. has nothing to disclose. G.W. has nothing to disclose. C.E.M.T.v.B. has nothing to disclose. C.B.L. has nothing to disclose. G.W.M. has nothing to disclose. D.I.B. has nothing to disclose.
This study was supported by the National Institute of Child Health and Human Development (HD042157); Center for Neurogenomics and Cognition Research (CNCR) VU University Amsterdam; the Twin-Family Database for Behavior Genetics and Genomics Studies (NWO-MagW 480-04-004) and Spinozapremie (NWO/SPI 56-464-14192). G.W.M is a National Health and Medical Research Council (NHMRC) Senior Research Fellow (grant no. 339446).
Reprint requests: Gonneke Willemsen, Department of Biological Psychology, VU University Amsterdam, van der Boechorststraat 1, 1081BT Amsterdam, The Netherlands (FAX: 31-20-5988832; E-mail: ahm.willemsen@psy.vu.nl).

and found that increasing height and BMI were related to an increased risk of spontaneous DZ twinning (adjusted odds ratios for being in the tallest quartile for body height and for being overweight were 1.66 and 2.07, respectively). Such an association was not found for MZ twinning. With respect to smoking, significantly higher multiple birth rates have been reported for mothers who smoke (17–20), although this association is not always significant (21).

Additional research is needed to verify the role of body composition and smoking in DZ twinning. We collected data from mothers of spontaneous DZ twins and mothers of spontaneous MZ twins on body composition and smoking history. Rather than using singletons as the control group, as was done in earlier studies, we compared mothers with spontaneous DZ twins to mothers with spontaneous MZ twins. Since MZ twinning is likely a random event, MZ twin mothers are as appropriate for controls as are singleton mothers. They may actually be the preferred controls as both MZ and DZ twin mothers need to be able to carry the twin pregnancy to full term.

In the analyses of the data, we took the established DZ twinning risk factors of maternal age and gravidity into account. In addition, we took into account the possible association between socioeconomic status and DZ twinning. Smoking behavior and body composition are associated with socioeconomic status (22–27), and socioeconomic status has been suggested to be associated with DZ twining (1, 10, 28), although the direction of the association is as yet unclear. We thus compared DZ twin mothers to MZ twin mothers for body composition and smoking before the twin pregnancy, after controlling for the effects of maternal age, gravidity, and educational attainment.

MATERIALS AND METHODS

Participants

The Netherlands Twin Registry (NTR) collects longitudinal data on twins and their family members in two subsamples: [1] newborn or young twins (YNTR) who are registered at birth by their parents and [2] adolescent and adult twins (ANTR) who self-register, along with their parents, siblings, and spouses (29, 30). About 90% of the participants are born in the Netherlands. A questionnaire was mailed to all mothers of twins (n = 33,528) registered with the NTR. Some of the mothers are passively registered, which means that they never participated in previous research projects. This was more often the case within the ANTR since initially addresses for these twin families were received through civil service administrations and may not always have been correct (29). From the group of actively registered mothers (n = 25,620), we received 17,683 completed questionnaires, and 1674 completed questionnaires were received from mothers who had never participated before. In total, 19,357 twin mothers participated in this study. The average time between the birth of the twins and the survey was 10.4 years (SD = 7.7).

Body Height, BMI at Time of Twin Pregnancy, and Smoking before Twin Pregnancy

The mothers were asked about their height and to indicate their weight just before the twin pregnancy. Data on height were divided into four quartiles (141–165, 165–169, 170–173, 174–195 cm). BMI before the pregnancy was calculated as BMI = weight in kilograms/height in meters squared and categorized according to the same categories used by Basso et al. (15) and Reddy et al. (16): less than 20 (15–19.99, low), 20–24.99 (normal), 25–29.99 (overweight), and 30 or more (30–47.99, obese).

The mothers were also asked about their smoking behavior before and during the twin pregnancy. They could report their smoking behavior as [1] no; [2] yes, but only before the pregnancy; [3] yes, but only during the twin pregnancy; [4] yes before and during the twin pregnancy; or [5] I don't know. Smoking at any time before the twin pregnancy was divided into two categories, nonsmoker and smoker before the twin pregnancy. Mothers who reported having smoked only during the twin pregnancy (n = 31) were also classified as smokers.

Maternal Age, Gravidity, and Educational Attainment

Maternal age at time of the twin pregnancy was obtained by subtracting the birth date of twins from the birth date of the mother and was recoded into the same four categories as reported in by Basso et al. (15); 25 or younger (17–25), 26–30, 31–35, 36 or older (36–45).

Gravidity was defined as the number of pregnancies, including miscarriages, the mother had before the birth of the twins. Gravidity was divided into two categories: null (no pregnancies before the twin pregnancy) and ≥ 1 (one or more pregnancies before the twin pregnancy).

Information on educational attainment was not available from the questionnaire but was obtained from previous ANTR and YNTR survey data. To obtain a proxy for socioeconomic status of the family, the highest educational attainment level achieved within a family (by the father or the mother of the twins) was used. Educational attainment was classified into three categories: low (primary or lower secondary education), intermediate (higher secondary education), and high (college or university education).

Sample Selection

Of the 19,357 women who returned questionnaires, all mothers who were not the biological mothers of the twins (n = 97) were excluded.

In the questionnaire, the twin mothers were also asked about conception of the twin/multiple pregnancy. The answers were [1] spontaneously conceived (with a request for specification of the time it took to become pregnant; 0–2, 3–5, 6–12, or more than 12 months), [2] IVF, [3] intracytoplasmic sperm injection, [4] IUI, [5] ovulation induction, or [6] other (with a request for specification). We excluded mothers who conceived their twins after the use of fertility techniques (n = 4451) or if data about mode of conception were missing (n = 108).

Finally, we excluded twin mothers when the zygosity of their twin offspring was unknown (n = 523), resulting in a sample of 14,178 mothers of twin offspring.

Based on sex of the twin offspring, 3777 twins were classified as opposite-sex DZ twins. In the case of same-sex twin offspring (10,401), we used zygosity based on DNA polymorphism, when available (for 16% of the same-sex twins). When this information was not available, zygosity from previous survey questions (for 65% of the same-sex twins) was used. These survey questions regarding offspring zygosity asked whether the twins were alike in eye color, hair color, face color, and face form and whether the twins were sometimes mistaken for each other by their parents, other relatives, or strangers. Based on the answers to these questions, twin zygosity was determined. Previous studies have shown the correspondence between DNA and questionnaire zygosity determination to be high in the NTR: 93% in the YNTR sample (31) and 97% in the ANTR sample (32). When data from previous studies were not available, we used the zygosity as reported in the current questionnaire by the mother (for 19% of the same-sex twins), who was asked to indicate whether the zygosity of the twin pair was MZ, DZ, or unknown. A comparison of the zygosity of the same-sex twin pairs provided by the mothers in the current questionnaire and the zygosity obtained from previous questionnaires showed a high degree of agreement (91%, n = 5953).

The final sample consisted of 5663 mothers of spontaneous MZ twins and 8515 mothers of spontaneous DZ twins. The data set consisted of 13,966 mothers with one twin pair and 212 mothers with two or more sets of multiples. In the case of multiple twin pregnancies, we only included the data of one twin pregnancy. In most cases, this meant the data for the firstborn twin were used. In the cases in which data were available for both twins and the firstborn twin was MZ while the second-born twin was DZ, we used data for the second-born twin pregnancy (n = 25).

Information on Nonresponders

Nonresponse can limit the interpretation of data collected in questionnaire studies (33–36). As indicated, we approached 25,620 twin mothers who had participated in earlier NTR surveys. This information was used to compare responders (n = 17,683) and nonresponders (n = 7937) to the present questionnaire. In this analysis, we included both mothers of spontaneous twins and mothers of twins conceived after fertility treatment to determine whether this was a factor influencing nonresponse. In responders and nonresponders, we compared zygosity (MZ or DZ), height (141–165, 165–169, 170–173, 174—196 cm), BMI (15–19.99, 20–24.99, 25–29.99, 30–48.99 kg/m^2), smoking during the twin pregnancy (yes or no), age at the time of the twin pregnancy (17–25, 26–30, 31–35, 36–47), and highest educational attainment level of the mother and her spouse (low, intermediate, or high). Responders and nonresponders were also compared on the use of artificial fertility techniques to conceive the twins (yes or no); sib-ship size, that is, the number of siblings of the mother herself (no other siblings, one or two siblings, and three or more siblings); whether the mother herself is a twin (yes or no); whether other female family members who were mothers of twins were present (familial twinning, yes or no); religious status (not religious, religious but not active, or religious and active); and the urbanization level of the residence of the mother (very heavy to heavy, moderate, low to very low). The information for the comparison of responders and nonresponders came from earlier surveys that were collected over a 20-year period. The questions contained in the surveys sometimes changed over time or were only asked of YNTR or of ANTR mothers. The number of observations therefore differs for each of these variables (see Table 1).

Statistical Analyses

To compare responders and nonresponders, we performed Pearson χ^2-tests. When significant differences were found, we conducted a binary logistic regression analysis to test whether these significant associations applied to both zygosity offspring groups, that is, whether the response bias was the same for mothers of MZ and mothers of DZ twins.

To compare spontaneous DZ twin mothers with spontaneous MZ twin mothers, we used binary logistic regression. We tested a model with DZ twinning (defined as yes/no) included as the dependent variable and as independent variables for height, BMI, and smoking as well as age, gravidity, and educational attainment. The reference categories in this model were age at twin birth of 25 years or younger, low education, being nulliparous before the twin pregnancy, height less than 165 cm, normal BMI (20–24.9 kg/m^2) before twin pregnancy, and not having smoked before the twin pregnancy. In these analyses, data were only included when information was present for all variables entered in the model. Since educational attainment was obtained from previous surveys, this reduced the sample size substantially. We therefore also ran the model without educational attainment.

RESULTS

Comparison of Responders and Nonresponders

Table 1 summarizes the results for the comparison of responders and nonresponders to the questionnaire. Nonresponse status was associated with twin offspring zygosity (more MZ mothers participated), being shorter, smoking during the twin pregnancy, being younger at twin birth, and having a lower educational attainment. In addition, nonresponse was associated with having more brothers and sisters and living in an urbanized area. However, the response bias was similar in mothers of spontaneous MZ and DZ twins.

Association of Body Composition and Smoking before the Twin Pregnancy with DZ Twinning

Table 2 shows the distribution (% and number) of the variables of interest in mothers of spontaneous MZ and mothers of

TABLE 1

Characteristics of responders and nonresponders obtained from previous surveys (number of observations and % for each trait) and test results for response bias (χ^2 and *P*-value) and, when significant, for differences in response bias between mothers of MZ and DZ twins (logistic regression; OR, 95% CI, and *P*-value).

			Pearson χ^2		Logistic regression (DZ vs. MZ comparison)		
	Nonresponders (%)	Responders (%)	χ^2	*P*	OR	CI	*P*
Zygosity of the twins:			117.4	.00			
MZ	1505 (25.6)	5106 (33.3)					
DZ	4379 (74.4)	10,240 (66.7)					
Missing	2053	2337					
Height, cm:			36.7	.00			
141–165	2008 (29.8)	4066 (26.1)			Reference		
165–169	1461 (21.7)	3410 (21.9)			1.0	0.8–1.2	.85
170–173	1726 (25.6)	4171 (26.8)			0.9	0.7–1.1	.20
174–196	1541 (22.9)	3928 (25.2)			1.0	0.8–1.3	.73
Missing	1201	2108					
BMI, kg/m^2:			13.2	.00			
15–19.99	885 (13.4)	1949 (12.7)			Reference		
20–24.99	3735 (56.5)	8977 (58.3)			1.1	0.7–1.5	.79
25–29.99	1383 (20.9)	3226 (21.0)			1.1	0.8–1.4	.63
30–48.99	610 (9.2)	1228 (8.0)			0.8	0.6–1.1	.22
Missing	1324	2303					
Smoking during the twin pregnancy:			291.8	.00			
Yes	2211 (29.1)	3292 (19.3)			1.0	0.8–1.1	.69
No	5386 (70.9)	13,771 (80.7)			Reference		
Missing	340	620					
Maternal age at twin birth:			185.3	.00			
17–25	1231 (16.1)	1788 (10.3)			Reference		
26–30	3052 (39.3)	6819 (39.1)			1.1	0.8–1.4	
31–35	2649 (34.4)	6837 (39.2)			1.1	0.8–1.4	
36–45	787 (10.2)	1981 (11.4)			1.0	0.8–1.3	
Missing	232	226					
Educational attainment:			547.2	.00			
Low	1694 (39.6)	3046 (22.1)			Reference		
Intermediate	1527 (35.7)	5631 (40.9)			1.0	0.3–1.4	.58
High	1059 (24.7)	5106 (37.0)			1.1	0.7–1.1	.57
Missing	3657	3900					
Use of artificial fertility techniques:			0.0	.97			
Yes	1004 (16.6)	2392 (16.6)					
No	5058 (83.4)	12,031 (83.4)					
Missing	1875	3260					
Sib-ship size mother:			52.1	.00			
No other siblings	164 (3.6)	485 (3.5)			Reference		
One or two siblings	2281 (50.7)	7786 (56.7)			0.7	0.4–1.1	.08
Three or more siblings	2060 (45.7)	5459 (39.8)			1.1	0.9–1.2	.51
Missing	3432	3953					

Hoekstra. Body composition, smoking, and twinning. Fertil Steril 2010.

TABLE 1

Continued.

			Pearson χ^2		Logistic regression (DZ vs. MZ comparison)		
	Nonresponders (%)	Responders (%)	χ^2	*P*	OR	CI	*P*
Self part of twin:			0.6	.45			
Yes	129 (2.6)	354 (2.4)					
No	4837 (97.4)	14,354 (97.6)					
Missing	2971	2975					
Familial twinning:			1.1	.30			
No other twins	2063 (45.3)	6173 (44.5)					
Other twins	2487 (54.7)	7713 (55.5)					
Missing	3387	3797					
Religious status:			4.8	.09			
Not religious	1357 (35.2)	4517 (33.4)					
Religious, not active	1664 (43.2)	5916 (43.8)					
Religious and active	833 (21.6)	3076 (22.8)					
Missing	4083	4174					
Urbanization level:			57.2	.00			
(Very) heavy	2351 (30.2)	4495 (25.9)			Reference		
Moderate	1804 (23.2)	3990 (23.0)			1.0	0.8–1.1	.57
(Very) low	3627 (46.6)	8844 (51.0)			1.0	0.9–1.2	.90
Missing	155	354					

Note: Data on (non) responders were available from earlier surveys. Not all variables were collected in all surveys.

Hoekstra. Body composition, smoking, and twinning. Fertil Steril 2010.

spontaneous DZ twins who took part in the questionnaire study. Because data on education came from earlier surveys, the sample size is reduced to 10,719 for this variable. Table 3 presents the odds ratios (ORs) and 95% confidence intervals (CIs) for changes in the DZ/MZ twinning proportion. Results for the full model are presented on the left side of the table (n = 10,234). DZ twinning was associated with body composition. Mothers in the tallest quartile for height were 1.6 times as likely to have DZ twins versus MZ twins when compared with mothers who were in the shortest height quartile. Compared with mothers with a normal BMI, mothers who were overweight or obese were 1.3 and 1.2 times more likely to have DZ twins than MZ twins. Having a BMI lower than 20 reduced the chance of having DZ twins. Smoking before pregnancy was also associated with DZ twinning; twin mothers who had smoked before the twin pregnancy had an increased chance of being a DZ twin mother (OR = 1.4). In these comparisons, the possible effects of maternal age, gravidity, and educational attainment on DZ twinning are accounted for. As can be seen in Table 3, maternal age and gravidity were, as expected, associated with DZ twinning, but educational attainment was not. Since educational attainment was not available for all participants and was not significantly associated with DZ twinning, we reran the analysis excluding educational attainment from the model (n = 13,346). The results of this analysis, presented at the right side of Table 3, are similar to those for the model including educational attainment.

DISCUSSION

This study investigated whether DZ twinning is associated with maternal body composition and maternal smoking history. After taking into account maternal age at twin birth, gravidity, and educational attainment, mothers of DZ twins were significantly taller, had a higher BMI, and smoked more often before the twin pregnancy than mothers of MZ twins. As expected, increased maternal age and increased gravidity were significantly associated with spontaneous DZ twinning, but there was no association between educational attainment and spontaneous DZ twinning.

This study replicates previous findings on height, BMI, and smoking in relation to DZ twinning (1, 7, 10, 15, 16, 18, 20, 21). Our study further extends these findings by taking into account possible influences of socioeconomic status, which was operationalized as educational attainment. Educational attainment did not alter the association of body composition and smoking before the twin pregnancy with DZ twinning. In fact, while both positive (28) and negative (1) associations have been reported for socioeconomic status and twinning, our study did not show any association between educational attainment and DZ twinning.

We looked at the possibility that nonresponse bias affected the results. Nonresponse may influence results based on data collected in questionnaire research, although generally only

TABLE 2

Prevalence (number and %) of maternal characeristics of mothers of spontaneous MZ and DZ twins.

	n	Mothers of spontaneous MZ twins	Mothers of spontaneous DZ twins
Height, cm:			
141–164	3659	1646 (29.3)	2013 (23.8)
165–169	3089	1313 (23.4)	1776 (21.0)
170–173	3832	1480 (26.3)	2352 (27.9)
174–195	3485	1183 (21.0)	2302 (27.3)
Missing	113	41	72
BMI before twin pregnancy, kg/m^2:			
15–19.99	2004	872 (15.8)	1132 (13.7)
20–24.99	8625	3566 (64.4)	5059 (61.1)
25–29.99	2431	829 (15.0)	1602 (19.3)
30–47.99	758	266 (4.8)	492 (5.9)
Missing	360	130	230
Smoking before twin pregnancy:			
Nonsmoker	9158	3829 (67.9)	5329 (63.0)
Smoker	4949	1813 (32.1)	3136 (37.0)
Missing	71	21	50
Maternal age at twin birth:			
17–25	1519	745 (13.2)	774 (9.1)
26–30	5522	2359 (41.8)	3163 (37.2)
31–35	5539	2011 (35.6)	3528 (41.5)
36–45	1565	535 (9.5)	1030 (12.1)
Missing	33	13	20
Gravidity:			
Null	5360	2330 (42.2)	3030 (36.7)
$\geq$1	8412	3189 (57.8)	5223 (63.3)
Missing	406	144	262
Educational attainment:			
Low	2328	975 (21.2)	1353 (22.1)
Intermediate	4334	1893 (41.1)	2441 (39.9)
High	4057	1740 (37.8)	2317 (37.9)
Missing	3459	1055	2404

Hoekstra. Body composition, smoking, and twinning. Fertil Steril 2010.

small differences between responders and nonresponders have been found (36–39). Because data from earlier survey collections were available for a large proportion of nonresponders in this study, we could compare characteristics of both groups as a function of the zygosity of their offspring. These analyses demonstrated small but significant differences between responders and nonresponders. However, the same differences were seen in mothers of MZ and DZ twins. It is therefore unlikely that response bias influenced the comparisons between MZ and DZ twin mothers.

The present study reports on the comparison of mothers of spontaneous DZ twins with mothers of spontaneous MZ twins. The results may therefore be limited to multiple births, and results may be more pronounced when comparing DZ twin mothers with singleton mothers. However, MZ twinning is generally not found to be influenced by genetic, maternal, or environmental factors. If, as proposed, MZ twinning is a randomly occurring event, MZ twin mothers are just as appropriate for controls as singleton mothers. In fact, they may form an even better control group. Although the mechanisms leading to an MZ twin pregnancy are very different from the mechanisms leading to a DZ twin pregnancy (the focus of our study), both MZ and DZ twin mothers have to carry the twin pregnancy to full term.

We analyzed data from mothers after a spontaneous twin pregnancy. Raj and Morley (40) suggested that parents of twins are not very willing to answer questions regarding the mode of conception. However, in the present study, the percentage of missing data on this question was low (<1%), and van Beijsterveldt and colleagues (41) showed

TABLE 3

ORs and 95% CIs for the comparison of the proportion of DZ to MZ mothers in each age, gravidity, height, BMI, smoking, and educational attainment category. The left side and right side of the table give results for models including and excluding educational attainment.

	n	OR	95% CI	*P*	n	OR	95% CI	*P*
Height, cm:								
141–165	2699	Reference			3446	Reference		
165–169	2318	1.2	1.0–1.3	.00	2934	1.1	1.0–1.2	.02
170–173	2745	1.3	1.2–1.5	.00	3640	1.3	1.2–1.5	.00
174–195	2472	1.6	1.4–1.8	.00	3326	1.6	1.5–1.8	.00
BMI before twin pregnancy:								
15–29.99	1523	0.9	0.8–1.0	.09	1935	0.9	0.8–1.0	.08
20–24.99	6521	Reference			8331	Reference		
25–29.99	1707	1.3	1.1–1.4	.00	2352	1.4	1.2–1.5	.00
30–47.99	483	1.2	1.0–1.5	.03	728	1.3	1.1–1.5	.00
Smoking before twin pregnancy:								
Nonsmoker	6599	Reference			8637	Reference		
Smoker	3635	1.4	1.3–1.5	.00	4709	1.3	1.2–1.4	.00
Maternal age at twin birth:								
17–25	1098	Reference			1408	Reference		
26–30	4220	1.4	1.2–1.6	.00	5249	1.3	1.2–1.5	.00
31–35	3890	1.7	1.5–2.0	.00	5244	1.7	1.5–1.9	.00
36–45	1026	1.9	1.5–2.2	.00	1445	1.8	1.6–2.1	.00
Gravidity:								
Null	4042	Reference			5220	Reference		
≥1	6192	1.1	1.0–1.2	.00	8126	1.2	1.1–1.3	.00
Educational attainment:								
Low	2176	Reference					Excluded	
Intermediate	4153	0.9	0.8–1.0	.12			Excluded	
High	3905	0.9	0.8–1.0	.12			Excluded	

Note: The analysis was based on data for 10,234 persons for the model including educational attainment and based on data for 13,346 persons for the model excluding educational attainment.

Hoekstra. Body composition, smoking, and twinning. Fertil Steril 2010.

that the pattern of missing for this variable is not different from the pattern of missing for other traits such as height. Also, if women who received fertility treatment are less likely to respond to surveys including this question, we would have expected to find a response bias for this variable. As shown in Table 1, women who received fertility treatment were as likely to participate in the survey as women who conceived spontaneously. A related issue is whether women accurately report on the mode of conception. Using a subset of the NTR sample, van Beijsterveldt and colleagues compared data on self-reported mode of conception with the data from hospital records (41) and found that mothers accurately report on the mode of conception.

The data were obtained through self-report after the twin pregnancy, on average 10 years later. Although height in this age group is a trait that remains stable across this time period, weight and smoking behavior may have changed in this time period, and recall may have influenced the results. Should either the actual change or the recall be different in MZ and DZ twin mothers, this would bias the results. However, when we performed the analysis separately for the ANTR and YNTR (older and younger cohort), the results for BMI and smoking were in the same direction. In the ANTR (n = 1486), being overweight increased the chance of having DZ twins by 1.9 (95% CI, 1.3–2.8), and having smoked before the pregnancy increased the chance by 1.1 (95% CI, 0.9–1.4), while in the YNTR (n = 11,860) these odds were 1.4 (95% CI, 1.2–1.6) and 1.4 (95% CI, 1.3–1.5), respectively.

For smoking behavior before the twin pregnancy, the way the question was phrased may have influenced the results. Smoking status was defined based on a four-category variable, which roughly identified persons as having smoked or not having smoked before the twin pregnancy. The option

"yes, smoked during the pregnancy but not before" was answered by 31 twin mothers, and they were classified as smokers before the pregnancy. It is possible that these mothers were misclassified, although due to the small number, the effect would be negligible. No information was obtained on smoking duration or number of cigarettes smoked. In addition, there was no specification of the time period between the twin pregnancy and prior smoking. When we compared our classification as smoker/non-smoker prior to twin birth with the data on smoking prior to the twin pregnancy (yes/no) from the questionnaire that mothers completed within the first year after twin birth (available for 11,971 twin mothers), a correspondence of 93.5% was found.

Similarly, educational attainment may reflect the education level obtained after the twin pregnancy. However, the vast majority of the sample was 26 years or older at the time of the twin pregnancy, and therefore most mothers will have reached their highest educational attainment. The average age at which women in the Netherlands have their first child has increased over the last 30 years predominantly owing to the fact that women will wait to have their first child until they have finished their education (42). Here too a comparison with an earlier survey in the younger cohort that was nearer to the birth of the twins (YNTR data collection took place when twins were 3 years old) showed similar results to those for the total sample. Although some bias has occurred, it is likely to be small and to occur in both MZ and DZ mothers and is thereby unlikely to influence the observed differences between MZ and DZ twin mothers.

Overall, our results extend previous findings that smoking and body composition are related to DZ twinning. Surprisingly, the same factors we observed to be associated with spontaneous DZ twinning are also associated with fertility problems. Maternal age, obesity, and smoking are all associated with an increased risk of infertility (43–46). The relationships between these factors are likely to be complex, and it is unclear whether effects on twinning or fertility act through the same or different mechanisms.

In conclusion, DZ twinning is moderately but significantly associated with body composition and smoking. Other mechanisms including genetic factors are likely to play a role, and it is also possible that these factors interact with body composition and smoking in increasing the risk of having DZ twins.

REFERENCES

1. Nylander PP. The factors that influence twinning rates. Acta Genet Med Gemellol (Roma) 1981;30:189–202.
2. Bulmer MG. The Biology of Twinning in Man. Oxford: Oxford Clarendon Press, 1970.
3. Derom R, Orlebeke J, Eriksson A, Thiery M. The epidemiology of multiple births in Europe. In: Keith LG, Papiernik E, Keith DM, Luke B, eds. Multiple Pregnancy. 1st ed. London: Parthenon Publishing Group, 1995: 145–62.
4. Hoekstra C, Zhao ZZ, Lambalk CB, Willemsen G, Martin NG, Boomsma DI, et al. Dizygotic twinning. Hum Reprod Update 2008;14: 37–47.
5. Statistics Netherlands. Geboorte naar diverse kenmerken. Centraal Bureau voor de Statistiek, http://statline.cbs.nl/StatWeb/start.asp?LA=nl&DM=SLNL&lp=Search/Search; 2007.
6. Tong S, Caddy D, Short RV. Use of dizygotic to monozygotic twinning ratio as a measure of fertility. Lancet 1997;349:843–5.
7. Bortolus R, Parazzini F, Chatenoud L, Benzi G, Bianchi MM, Marini A. The epidemiology of multiple births. Hum Reprod Update 1999;5: 179–87.
8. Hall JG. Twinning. Lancet 2003;362:735–43.
9. Obi-Osius N, Misselwitz B, Karmaus W, Witten J. Twin frequency and industrial pollution in different regions of Hesse, Germany. Occup Environ Med 2004;61:482–7.
10. MacGillivray I, Samphier M, Little J, Ian M, Dorris MC, Barbara T. Factors affecting twinning. In: MacGillivray I, Campbell DM, Thompson B, eds. Twinning and Twins. New York: John Wiley and Sons, 1988:67–97.
11. Eriksson AW, Fellman J. Seasonal variation of livebirths, stillbirths, extramarital births and twin maternities in Switzerland. Twin Res 2000;3: 189–201.
12. Martin JA, Hamilton BE, Sutton PD, Ventura SJ, Menacker F, Munson ML. Births: final data for 2003. Natl Vital Stat Rep 2005;54: 1–116.
13. Fauser BC, Devroey P, Macklon NS. Multiple birth resulting from ovarian stimulation for subfertility treatment. Lancet 2005;365:1807–16.
14. Lambalk CB, Schats R, Bleker OP, Elfering-Stinkens PM, Orlebeke JF. Meerlingzwangerschappen; epidemiologie en beleid. Ned Tijdschr Geneeskd 2004;148:448–50.
15. Basso O, Nohr EA, Christensen K, Olsen J. Risk of twinning as a function of maternal height and body mass index. JAMA 2004;291:1564–6.
16. Reddy UM, Branum AM, Klebanoff MA. Relationship of maternal body mass index and height to twinning. Obstet Gynecol 2005;105:593–7.
17. Yerushalmy J. Mother's cigarette smoking and survival of infant. Am J Obstet Gynecol 1964;88:505–18.
18. Olsen J, Bonnelykke B, Nielsen J. Tobacco smoking and twinning. Acta Med Scand 1988;224:491–4.
19. Kallen K. Maternal smoking and twinning. Twin Res 1998;1:206–11.
20. Morales-Suarez-Varella MM, Bech BH, Christensen K, Olsen J. Coffee and smoking as risk factors for twin pregnancies. The Danish National Birth Cohort. Twin Res Hum Genet 2007;10:597–603.
21. Parazzini F, Chatenoud L, Benzi G, Di Cintio E, Dal Pino D, Tozzi L, et al. Coffee and alcohol intake, smoking and risk of multiple pregnancy. Hum Reprod 1996;11:2306–9.
22. Conrad KM, Flay BR, Hill D. Why children start cigarettes: predictors of onset. Br J Addict 1992;87:1711–24.
23. Ericson A, Gunnarskog J, Kallen B, Otterblad-Olausson P. Surveillance of smoking during pregnancy in Sweden, 1983–1987. Acta Obstet Gynecol Scand 1991;70:111–7.
24. Laaksoonen M, Rahkonen O, Prattala R. Smoking status and relative weight by educational level in Finland, 1978–1995. Prev Med 1998;27:431–7.
25. Willemsen MC, Hoogeveen RT, van der Lucht F. New smokers and quitters. Transitions in smoking status in a national population. Eur J Public Health 2002;12:136–8.
26. Ball K, Mishra G, Crawford D. Which aspects of socioeconomic status are related to obesity among men and women? Int J Obes Relat Metab Disord 2002;26:559–65.
27. Martinez JA, Kearney JM, Kafatos A, Paquet S, Martinez-Gonzalez MA. Variables independently associated with self-reported obesity in the European Union. Public Health Nutr 1999;2:125–33.
28. Bonnelykke B. Social class and human twinning. J Biosoc Sci 1990;22: 381–6.
29. Boomsma DI, de Geus E, Vink JM, Stubbe JH, Distel MA, Hottenga JJ, et al. Netherlands Twin Register from twins to twin families. Twin Res Hum Genet 2006;9:849–57.
30. Bartels M, van Beijsterveldt CEM, Derks EM, Stroet TM, Polderman JC, Hudziak JJ, et al. Young Netherlands Twin Register (Y-NTR): a longitudinal multiple informant study of problem behavior. Twin Res Hum Genet 2007;10:3–11.

31. Rietveld MJ, van der Valk JC, Bongers IL, Stroet TM, Slagboom PE, Boomsma DI. Zygosity diagnosis in young twins by parental report. Twin Res 2000;3:134–41.
32. Willemsen G, Posthuma D, Boomsma DI. Environmental factors determine where the Dutch live: results from the Netherlands twin register. Twin Res Hum Genet 2005;8:312–7.
33. Barchielli A, Balzi D. Nine-year follow-up of a survey on smoking habits in Florence (Italy): Higher mortality among non-responders. Int J Epidemiol 2002;31:1038–42.
34. Goyder J, Warriner K, Miller S. Evaluating socio-economic status (SES) bias in survey nonresponse. J Off Stat 2002;18:1–11.
35. Korkeila K, Suominen S, Ahvenainen J, Ojanlatva A, Rautava P, Helenius H, et al. Non-response and related factors in a nation-wide health survey. Eur J Epidemiol 2001;17:809–17.
36. Van Loon AJM, Tijhuis M, Picavet HSJ, Surtees PG, Ormel J. Survey non-response in the Netherlands: effects on prevalence estimates and associations. Ann Epidemiol 2003;13:105–10.
37. Vink JM, Willemsen G, Stubbe JH, Middeldorp CM, Ligthart RS, Baas KD, et al. Estimating non-response bias in family studies: application to mental health and lifestyle. Eur J Epidemiol 2004;19: 623–30.
38. Distel MA, Ligthart L, Willemsen G, Nyholt DR, Trull TJ, Boomsma DI. Personality, health and lifestyle in a questionnaire family study: a comparison between highly cooperative and less cooperative families. Twin Res Hum Genet 2007;10:348–53.
39. Heath AC, Howells W, Kirk KM, Madden PA, Bucholz KK, Nelson EC, et al. Predictors of non-response to a questionnaire survey of a volunteer twin panel: findings from the Australian 1989 twin cohort. Twin Res 2001;4:73–80.
40. Raj S, Morley R. "Are you asking me if we had sex to conceive?" To whom do parents of twins disclose mode of conception and what do they feel about being asked? Twin Res Hum Genet 2007;10:886–91.
41. Van Beijsterveldt CEM, Hoekstra C, Schats R, Montgomery GW, Willemsen G, Boomsma DI. Mode of conception of twin pregnancies: willingness to reply to survey items and comparison of survey data to hospital records. Twin Res Hum Genet 2008;11:349–51.
42. van Agtmaal-Wobma E, van Huis M. De relatie tussen vruchtbaarheid en opleidingsniveau van de vrouw. [The relationship between fertility and educational attainment of the woman]. Bevolkingtrends 2008;56 2e kwartaal:32–41.
43. Flegal KM, Carroll MD, Ogden CL, Johnson CL. Prevalence and trends in obesity among US adults, 1999–2000. JAMA 2002;288:1723–7.
44. Goumenou AG, Matalliotakis IM, Koumantakis GE, Panidis DK. The role of leptin in fertility. Eur J Obstet Gynecol Reprod Biol 2003;106: 118–24.
45. Hedley AA, Ogden CL, Johnson CL, Carroll MD, Curtin LR, Flegal KM. Prevalence of overweight and obesity among US children, adolescents, and adults, 1999–2002. JAMA 2004;291:2847–50.
46. Lash MM, Armstrong A. Impact of obesity on women's health. Fertil Steril. Published online 14 April 2008. [E-pub ahead of print.]