EFFECTS OF PRENATAL NICOTINE EXPOSURE ON INFANT LANGUAGE DEVELOPMENT: A COHORT FOLLOW UP STUDY

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ABSTRACT

Background: To study the longitudinal effects of prenatal nicotine exposure on cognitive development, taking into consideration prenatal and postnatal second-hand smoke exposure.

Methods: A cohort follow up study was carried out. One hundred and fifty-eight pregnant women and their infants were followed during pregnancy and infant development (at 6, 12 and 30 months). In each trimester of pregnancy and during postnatal follow-up, a survey was administered to obtain sociodemographic data and the details of maternal and close familial toxic habits. Obstetric and neonatal data were obtained from hospital medical records. To assess cognitive development, the Bayley Scales of Infant Development were applied at 6, 12 and 30 months; to assess language development, the MacArthur-Bates Communicative Development Inventories were applied at 12 months and the Peabody Picture Vocabulary Test at 30 months.

Results: After adjustment for confounding variables, the results showed that infants prenatally exposed to cigarette smoke recorded poor cognitive development scores. Language development was most consistently affected, specifically those aspects related to auditory function (vocalizations, sound discrimination, word imitation, prelinguistic vocalizations, and word and sentence comprehension).

Conclusions: Irrespective of prenatal, perinatal and sociodemographic data (including infant postnatal nicotine exposure), prenatal exposure to cigarette smoke and second-hand smoke affect infant cognitive development, especially language abilities.

SIGNIFICANCE

What is already known? It is already known that maternal smoking during pregnancy is associated to behavioral and cognitive alterations in offspring. These aspects are also related to other prenatal and postnatal factors such emotional states during pregnancy, obstetrical and perinatal conditions, sociodemographical factors, and postnatal infant secondhand smoke.

What this study adds? This study offers new longitudinal data (pregnancy, 6, 12 and 30 months old) about the effects of maternal smoking during pregnancy on infant cognitive development, and concretely, on infant language development. These relationships have been studied taking into account a group of prenatal secondhand exposed infants; and controlling for a series of confounding prenatal and postnatal variables: prenatal maternal anxiety, neonatal birth weight, gestational age at birth, parity, postnatal exposure to tobacco smoke, infant breastfeeding, family socioeconomic status (SES), parents' general psychopathology and mother-infant attachment.

INTRODUCTION

There is clear evidence that maternal smoking during pregnancy (MSDP) has a bearing on neonatal morbidity; nevertheless, tobacco is the most widely used toxic substance during pregnancy. Epidemiological studies have shown that between 11 and 30% of pregnant women smoke or are exposed to secondhand smoke (SHS), and this rate increases to 50% in high-risk samples, including young, poor and urban populations. Even so, in many industrialized countries, the rates of women who actively smoke appear to have peaked and have now begun to decline (Mathews, 2001).

MSDP is consistently related to various health problems and behavioral and cognitive impairments in offspring(Melo, Bellver, & Soares, 2012). MSDP and SHS exposure are related to increased irritability and excitability, alterations in crying behavior and soothability, decreased alertness, and low neonatal maturity (Hernández-Martínez, Arija Val, Escribano Subías, & Canals Sans, 2012). These problems persist throughout the life of the child, manifesting as behavioral problems such as negativity, difficult temperament, attention disorders, hyperactivity and aggressiveness(Button, Maughan, & McGuffin, 2007). A specific effect of MSDP on cognitive development in infants and children has also been described. In this sense, MSDP have been related to delayed psychomotor and mental scores in 2-year-old preterm infants (Kiechl-Kohlendorfer et al., 2010), to poor motor and cognitive development in 13-months-old to 12-year-old children (Sexton, Fox, & Hebel, 1990), and to poor performance in specific cognitive areas such as sustained attention, working and design memory, arithmetic tasks and problem solving in 4 to 11-year-old children (Batstra, Hadders-Algra, & Neeleman, 2003; Julvez et al., 2007). Specific deficits in language-related abilities were also observed. One of the most consistent findings is an association between MSDP and lower performance in spelling tasks, specific language and auditory tests, reading and language performance, verbal learning, receptive language, central auditory processing and visual perceptual processing (Batstra et al., 2003; Fried,

Watkinson, & Gray, 1992; Fried & Watkinson, 2000; Mccartney, Fried, & Watkinson, 1994). These effects appear to persist to at least 12 years of age (Fried & Watkinson, 2000). SHS exposure during pregnancy and cognitive development is a condition less studied. Lee et al. (2011) found that 6-month-old prenatally exposed to SHS infants had an increased risk to have developmental delay.

Nicotine can affect fetal brain development in several ways. First, tobacco smoke interferes with normal placental function, acting as a vasoconstrictor that reduces uterine blood flow to the fetus, impeding oxygen and nutrient transport. This compromises fetal intrauterine growth and central nervous system development (Roos et al., 2015). Second, nicotine is a neuroteratogen that can cross the placenta and target brain nicotinic acetylcholine receptors in critical developmental periods, changing the pattern of cell proliferation, differentiation and myelination (Dwyer, Broide, & Leslie, 2008); and the third way by which MSDP affects infant development is the fetal programming way. Prenatal smoke exposure certainly contributes to creating an adverse fetal environment that affects programming, for example, MSDP has been associated with atypical DNA methylation patterns in brain-derived neurotrophic factor (BDNF) and dysregulated expression of microRNA; however, studies considering epigenetic pathways have only recently begun to emerge (Knopik, Maccani, Francazio, & McGeary, 2012).

In addition to smoking during pregnancy there are other factors that have a bearing on child cognitive development which include maternal education and psychopathology, obstetrical conditions, mother-infant attachment, infant breastfeeding, etc. (Conroy et al., 2012; Ding, Xu, Wang, Li, & Wang, 2014; Koutra et al., 2012; Victora et al., 2015). There are postnatal conditions that can modify the adverse effects of MSDP on a child's cognitive development. Batstra et al. (Batstra, 2003) and Obel et al. (Obel, Henriksen, Hedegaard, Secher, & Østergaard, 1998) found that the negative effects of MSDP on babbling behavior at 8-month-old; and on reading, spelling and arithmetic tasks at 9 years old were found in those children who had not been breastfed or were breastfed for less than four months. Postnatal SHS exposure seems to increase the effect of MSDP, in this sense Eskenazi and Castorina (Eskenazi & Castorina, 1999) found in their revision that postnatal SHS exposure were often associated with impaired development. All these correlations make causal attribution difficult. Longitudinal studies with repeated measures of neurobehavioral deficits across key developmental periods must be performed to obtain a valid epidemiological report, and numerous covariates must be controlled in order to disentangle the compound effects statistically from the demographic background, maternal psychosocial status, paternal characteristics, other substance use, perinatal

conditions and environmental factors (Melo et al., 2012). Consequently, the aim of this study is to examine the effect of prenatal smoke exposure on infant cognitive development at 6, 12 and 30 months old, taking into account a group of mother exposed to SHS and controlling for a series of confounding prenatal and postnatal variables: prenatal maternal anxiety, neonatal birth weight, gestational age at birth, parity, postnatal exposure to tobacco smoke, infant breastfeeding, family socioeconomic status (SES), parents' general psychopathology and mother-infant attachment. We hypothesize that, (H1) independently of prenatal and postnatal confounding variables, infants prenatally exposed to MSDP and SHS will have poor performance in cognitive development scales than infants not exposed; and that (H2) these poor scores will be done along the follow up.

METHODS

Sample

The sample size required for the present study was determined to be 119 mother-father-infant triads distributed in the three groups of prenatal smoking habits. The estimation of this sample size was based on: an expected mean difference between groups of 10 points, a 5% of alpha one-sided error and on a statistical power of 90%. At 6 and 12 months, the sample was 156 and 134 respectively; and according to the means in the cognitive scores and distribution on smoking groups, the final statistical power was of 95%. At 30 months, the sample was 92, and according to the above mentioned parameters, the final statistical power was of 80%.

Participants were pregnant women recruited during the first trimester pregnancy and their infants. Recruitment was carried out over the period 2005–2009 by obstetricians and pediatricians at Sant Joan University Hospital in Reus (Spain). The eligibility criteria for pregnant women were to be pregnant, over 18 years of age, at no more than 11 weeks of gestation, being healthy, with a singleton pregnancy, and having no chronic illness affecting nutritional status, such as diabetes type I, Crohn's disease and celiac disease. The inclusion criteria for children were to be born in the Sant Joan University Hospital in Reus, at term, of normal weight, with no medical problems during the first days of life. There were no significant differences in perinatal, psychological and sociodemographic variables between subjects who completed the study and those who were excluded or dropped out along follow up. The number of included and excluded participants along the follow-up and the study design is shown in Figure 1.

Procedure and Study design

A cohort follow up study (DEFENSAS cohort study) was carried out.

The study was approved by the Research and Ethics Committee of Sant Joan University Hospital in Reus (Spain) and informed consent was obtained from participants. Pregnant women that met the study criteria and were willing to participate in the study were recruited by the hospital gynecologist during their first gynecology consultation. Newborns who met the study criteria were then included in the follow-up stage. The study design is shown in Figure 1.

Variables and instruments

Outcomes variables: Infant cognitive measures

The Bayley Scales for Infant Development (BSID)(Bayley, 1993) was used to assess cognitive development. The BSID is an individually administered examination that assesses the current developmental functioning of infants from 0 to 42 months old and consists of three scales: the mental scale (assesses memory, habituation, problem solving, early number concepts, generalization, classification, vocalizations, and language and social skills), the motor scale (assesses the control of the gross and fine muscle groups) and the behavior rating scale. From these scales, a mental development index (MDI) and a psychomotor development index (PDI) can be obtained. Moreover, with combined information from all items a developmental age for the cognitive, language, personal-social and motor areas can be also obtained. For the current study we used MDI, PDI, and ages of cognitive, language, personal-social and motor development. BSID was administered at 6, 12 and 30 months old at the hospital by three trained developmental psychologists. All caregivers were present during the assessments.

The MacArthur-Bates Communicative Development Inventories (Spanish Adaptation) (MBCDI)(López Ornat, S., Gallego, C., Gallo, P., Karousou, A., Mariscal, S., Martínez, 2005) are a parent assessment measure of children's early language development, comprising two forms: the *Words and Gestures* form, for children aged 8–15 months, and the *Words and Sentences* form, for children aged 16–30 months. The participating parents completed the *Words and Gestures* form, which provides a percentile measure of vocabulary comprehension, vocabulary production and use of gestures. The MBCDI had been administered to the parents when their child was 12-13 months old.

The Peabody Picture Vocabulary Test-Third Edition (Spanish adaptation) (PPVT-III)(Campbell, Bell, & Keith, 2001) is an individually administered examination that assesses receptive oral vocabulary in people aged from 30 months to 90 years old. It provides one total standard score (mean=100, SD=15) and can be used as an estimate of general verbal ability. The PPVT-III was administered to each child at 30 months old at the hospital.

Exposures: Smoking measures

Data on MSDP were collected using a survey specifically designed for this study, which was administered by a member of the research team at each trimester (weeks 11-12, 19-20 and 31-32) of gestation. Assurances of data confidentiality were given to encourage participants to provide candid responses. To collect data on prenatal toxic habits, participants were asked about their consumption of cigarettes, alcohol and other drugs. Smoking habits were determined by asking the following question: *Do you smoke?* If the response was negative, the pregnant woman was included in the non-smoking group; if the response was affirmative, we asked about current smoking habits and placed participants in one of five categories according to the number of cigarettes smoked per day: 1–5, 6–10, 11–15, 15–20 and >20. Second-hand smoke exposure was determined by asking the following questions: *Do you usually smell tobacco smoke at home? Do you usually smell tobacco smoke at your workplace?* Non-smoking pregnant women who responded affirmatively to either of these questions were included in the SHS exposure group. We also recorded whether the mother had quit smoking upon becoming pregnant. Twenty mothers reported having quit smoking before 9 weeks of gestation. A variance analysis was performed to examine the differences between non-smokers and women who quit smoking once pregnant; the results revealed that the maternal, infant and sociodemographic characteristics of mothers who had stopped smoking upon becoming pregnant and the outcome variable (infant cognitive development) for their newborns were comparable to those of the non-smoking group; therefore, these mothers were included in the non-smoking group.

Potential confounders: Adjustment measures

Data for obstetric and neonatal variables were collected from the medical records of each woman in the immediate postpartum period.

Data for infant postnatal smoke exposure were collected by personal interviews with parents during the infant assessments at 6, 12 and 30 months old. The researcher asked parents: *Do you smoke?* If the response was negative,

we included the infant in the no smoking exposure group, and if the response was affirmative, we asked: *Do you smoke at home? Do you smoke in the presence of your child?* If the responses were negative, the infant was placed in the no smoking exposure group, and if the response to either question was affirmative, the infant was included in the smoking exposure group.

Data for infant breastfeeding were collected during the infant assessments in the immediate postpartum and at age 6, 12 and 30 months.

Socioeconomic status (SES) data were obtained using the Hollingshead Four Factor Index of Socioeconomic Status(Hollingshead, 2011). For this study, we determined family SES by combining the data obtained from the father and the mother. Data were collected at 11-12 weeks of gestation.

The parents' general health status was determined using the Spanish version of the General Health Questionnaire (GHQ-28) (Lobo, Pérez-Echeverría, & Artal, 1986), a questionnaire that assesses psychiatric symptoms obtaining a total score of general distress. The GHQ-28 was administered to mothers at infant age of 6, 12 and 30 months, and to fathers at 30 months. If there were missing items and they not suppose more than 10% of the total, we imputed data according to the response of similar items.

Infant-mother attachment was assessed using the Parenting Stress Index (PSI) (Abidin, 1995), which is a parent report of the effect that parenting has upon an individual's stress level. The PSI offers scores of Child Domain (reinforces parent, mood, acceptability, adaptability, demandingness and distractibility/hyperactivity) and Parent Domain (depression, attachment, isolation, competence, spouse, role restriction, and health). For this study we used the "attachment" sub-domain collected at 6, 12 and 30 months old. If there were missing items and they not suppose more than 10% of the total, we imputed data according to the response of similar items.

Maternal anxiety was evaluated using the Spanish version of the State-Trait Anxiety Inventory (STAI)(Spielberger, Gorsuch, & Lushene, 1997) which is a questionnaire that assesses the state anxiety (the level of transient and situational anxiety) and trait anxiety (the level of dispositional and stable trait anxiety). For this study we used the trait anxiety score which was obtained in the immediate postpartum. There were no missing items.

Statistical analysis

ANOVA and Chi-Square tests were used to study the differences between prenatal smoking exposure groups in terms of obstetric, infant and sociodemographic variables.

To adjust the BSID-II, MBCDI and Peabody scores for potential confounders and covariates (child gender, maternal prenatal anxiety, birth weight, gestational age at birth, parity, SES, infant's age at cognitive assessment, infant postnatal smoke exposure, months of breastfeeding, mother-infant attachment and parent psychopathological and health general status), a linear regression analysis using the enter method was conducted. Previously, hypothesis of linearity was tested. Based on these analyses, adjusted BSID-II, MBCDI and Peabody scores were obtained. The differences between prenatal smoking exposure groups in the adjusted BSID-II, MBCDI and Peabody scores were analyzed using parametric (ANOVA) and non-parametric (Mann-Whitney) methods (depending upon the distribution of scores).

The scores that were not normally distributed were summarized by median and interquartile ranges.

Bonferroni correction was applied to control for the increase in type I error due to multiple comparisons; the significance level was 0.02.

Data were analyzed using SPSS Statistics Desktop 22.0.

RESULTS

Descriptive data of the sample

The descriptive data of the sample are shown in Table 1, and the scores of the cognitive test are shown in table 2. Mother's mean age at partum was 31.42 (SD=4.41), and a 58.2% were multiparous. A 51.9% of the infants were boys and all the infants were born healthy and at term. Regarding smoking habits, a 62.7% of the mothers did not smoke (20 quit smoking upon becoming pregnant), of the mothers exposed to smoke, 17.1% were exposed to SHS and 20.2% smoked during pregnancy. Of the infants exposed to smoke, 34.8% have a close relative who smokes at home.

Descriptive data of the sample in relation to prenatal smoking exposure

The obstetric, infant and sociodemographic characteristics of the sample by smoking groups are shown in Table 3. In terms of sociodemographic characteristics, non-smokers were significantly older (mean age=32.13; SD=3.80) than mothers exposed to SHS (mean age=29.48; SD=5.43) and smokers (mean age=30.88; SD=4.80) mothers. The data also show a significant relationship between SES and smoking habits during pregnancy in terms that in lower SES groups were more smokers and SHS exposed mothers.

Relationship between prenatal smoking exposure and infant cognitive development at 6, 12 and 30 months old

No significant differences were found before adjust cognitive variables for confounders. Table 4 shows differences between groups of prenatal smoking exposure in terms of BSID adjusted scores at 6, 12 and 30 months old, MBCDI adjusted scores at 12 months, and Peabody adjusted scores at 30 months. At 6 months old, infants prenatally exposed to SHS (mean=5.01; SD=0.41) and to smoking (mean=4.92; SD=0.43) scored significantly lower in the BSID-II for language development age (F=9.692; p=0.001) than infants not prenatally exposed to smoking (mean=5.40; SD=0.46).

At 12 months old, infants prenatally exposed to smoking (mean=89.32; SD=2.66) scored significantly lower in the BSID for the PDI (F=4.797; p=0.010) than infants not prenatally exposed (mean=91.41; SD=2.88). Likewise, infants prenatally exposed to SHS (mean=9.87; SD=0.28) obtained lower scores for language development age (F=5.142; p=0.007) than infants not prenatally exposed to smoking (mean=10.22; SD=0.31). For the MBCDI, infants prenatally exposed to SHS (mean=85.78; SD=2.45) and to smoking (mean=86.17; SD=2.55) scored lower than infants not exposed (mean=89.93; SD=2.53) for early comprehension (F=11.148; p=0.001) and word comprehension (F=8.662 (p=0.001); mean=51.02 (SD=4.52) for SHS exposed groups; mean=51.59 (SD=4.95) for smoking groups and mean=54.35 (SD=3.90) for non smoking groups). Moreover, significant differences were observed in the scores for prelinguistic vocalizations (F=3.666; p=0.020) between infants exposed to SHS (mean=67.65; SD=5.98) and infants not exposed to smoking (mean=71.34; SD=6.32), and in the scores for sentence comprehension (F=4.581; p=0.012) between infants exposed to smoking (mean=58.54; SD=3.44) and infants not exposed to smoking (mean=61.21; SD=3.36).

At the end of follow-up at 30 months old, infants exposed to smoking and SHS scored significantly lower than those not exposed to smoking in the BSID-II for the MDI (F=7.156, p=0.001; mean=97.08, SD=3.57; mean=98.15, SD=3.73

and mean=100.82, SD=3.86, respectively) and language developmental age (Chi=13.250, p=0.001; median=33.76, IQR=1.68; median=33.12, IQR=2.36 and median=35.00, IQR=1.98 respectively). For the PDI, infants prenatally exposed to SHS (mean=95.45; SD=4.35) scored significantly lower (F=6.729; p=0.002) than infants not exposed to smoking (mean=98.50; SD=3.70) and infants exposed to smoking (mean=98.21; SD=3.94). No significant differences between groups were observed for Peabody IQ scores.

DISCUSSION

Cognitive and language development is a complex process influenced by several factors. Our analysis included confounding variables that are related to it, such as obstetric outcomes, prenatal anxiety, family SES, mother-father-infant attachment and parents' general psychological health. We also took into account variables that can decrease the adverse effects of prenatal exposure to tobacco smoke, such as infant breastfeeding, and variables that can increase the adverse effect, such as postnatal exposure to tobacco smoke. These variables are considered important for future collaborative epidemiological research involving cognitive follow-up during infant development. Sociodemographic variables must also be taken into account, as they often show a correlation with smoking habits(Melo et al., 2012). In our sample, smoking behavior was associated with low SES and lower maternal age; to rest the confounding effect of these variables, both were included in the adjustment of cognitive scores.

In this 3-year follow up study, we found that MSDP and SHS are associated with poor cognitive development scores as we have hypothesized (H1 andH2). At 12 months old, infants prenatally exposed to SHS scored lower for the PDI than infants prenatally exposed to smoke, as previously suggested Kiechl-Kohelndorfer et al. (Kiechl-Kohlendorfer et al., 2010); in the long term, infants prenatally exposed to smoke and SHS scored lower for the MDI than infant not exposed. Our results showed no differences between these development indexes at 6 months, unlike Lee et al., (2011).

For language scores, our findings are more consistent and indicate that infants prenatally exposed to smoke and SHS showed poorer vocalization and gesture capabilities and are less able to discriminate familiar words at six months old. The same groups were less able to imitate words, to use words properly, to point to objects vocalized by the examiner, to produce prelinguistic vocalizations and to understand early words and sentences at 12 months old (although early language production is not affected). These results are consistent with Obel et al., (1998), who found

that 8-month-old infants prenatally exposed to smoke and SHS exhibit less babbling behavior. At 30 months old, our prenatal exposed group scored lower in receptive and expressive language capabilities, as found Mccartney et al. (1994). These results may reflect disturbances in auditory functioning that could cause alterations in language and verbal skills observed in older children (Fried & Watkinson, 2000; Mccartney et al., 1994). As has been previously stated, nicotine is a neuroteratogen that can cross the placenta and target fetal brain nicotinic acetylcholine (Dwyer et al., 2008) being the auditory pathway is heavily mediated by acetylcholine, so fetal nicotine exposure can upregulate nicotinic cholinergic receptor binding sites, causing abnormalities in the development of synaptic activity. In these sense, several studies have reported structural changes in brain regions related to auditory processing, which in turn may lead to the observed language difficulties. Jacobsen et al., (2007) reported structural alterations (maturational increases in cell packing density, fiber diameter, directional coherence and myelination) in anterior cortical white matter and the anterior limb of the internal capsule (which contain auditory fibers) in adolescents prenatally exposed to tobacco smoke. These alterations may indicate disruption of auditory nerve fiber growth, which causes deficits in auditory processing (Jacobsen et al., 2007). Functional alterations have been also described. Kable et al., (2009) analyzed the auditory brainstem responses of 6-month-old infants prenatally exposed to tobacco smoke and found that MSDP was negatively correlated with auditory brainstem response latency; the same higher latency was observed (together with poor discriminative capability) in the brainwave responses to different consonant-vowel syllables of 48-hour-old newborns prenatally exposed to tobacco smoke (Key et al., 2007). The results reflect a disruption in the sensory encoding of auditory stimuli that persists into middle childhood and is highly predictive of future reading and language difficulties.

Our data are relevant because there are few studies that assess these characteristics in young infants and that perform a longitudinal assessment across different periods of development, taking into account multiple confounders. We report results recorded at 6, 12 and 30 months old, but we also assessed the neonatal behavior of the sample at 48-72 hours and found that infants prenatally exposed to smoke and SHS already showed greater difficulty with the auditory habituation items of the NBAS (Hernández-Martínez et al., 2012). Even, fetuses exposed to maternal smoking were less responsive to the mother's voice (Cowperthwaite, Hains, & Kisilevsky, 2007).

One particular aspect that should be addressed is the timing with which the above effects are observed. The critical period for nicotine-induced brain damage appears to be the second and third trimester (Gatzke-Kopp & Beauchaine,

2007), when nicotinic receptors maximally influence neurodevelopment, with no effects noted when exposure is limited to early gestation. In our sample, the infants of mothers who quit smoking during the first trimester showed the same psychosocial, sociodemographic and cognitive characteristics as those of mothers not exposed to smoke, and were therefore included in the non-exposed group (Hernández-Martínez et al., 2012).

Our study has some limitations. The first limitation is the level of attrition during follow-up, which rises from 15.4% at 12 months to 37.2% at 30 months. This is a normal condition of follow-up studies, and there are no significant differences in sociodemographic, obstetric or pediatric data between those infants and families that underwent all of the assessments and those that dropped out. The second potential limitation is the measure of prenatal tobacco exposure used. We considered the mothers' responses to surveys to constitute valid reports, and each participant was asked about her toxic habits at seven different times to guarantee reliability (the first, second and third trimesters of gestation, immediate postpartum, and each follow-up visit); the research team treated all data confidentially, to encourage the participants to provide candid responses. While total reliability cannot be guaranteed, other authors have validated these types of surveys using saliva cotinine as a marker and their results suggest that self-reported smoking exposure during pregnancy is highly accurate (McDonald, Perkins, & Walker, 2005). Finally, cognitive development and later Intelligence Quotient (IQ) are clearly influenced by environment and genetics (Hansell et al., 2015), so, it would be desirable to adjust cognitive development by parents IQ. In this sense, we have used the SES variable which takes into account the parents academic level and the type of job which also correlates with IQ (Strenze, 2015).

Our results show that smoking exposure during pregnancy, even at low levels or through passive exposure, causes health, neurocognitive and behavioral problems in offspring and is therefore a public health concern. In Spain, public policies and laws were enacted to protect non-smokers and smoke is only permitted in the street and at home. Despite this progress, pregnant women and children may still be exposed to SHS at home, so, an extra effort must be done by clinicians (pediatricians, obstetricians and midwives) and education policymakers to encourage pregnant women to quit smoking before or early in pregnancy and to encourage other family members to refrain from smoking in shared spaces.

In conclusion, low levels of MSDP and maternal SHS exposure are related to infant cognitive development in the short and long term. Language development is most consistently affected, potentially leading to spelling, reading,

verbal learning and receptive language problems. As such, primary care physicians, obstetricians, pediatricians and health professionals in general should encourage mothers and close relatives to moderate their smoking habits, including them in smoking cessation programs and informing them of the effects of involuntary smoke exposure to prevent direct damage to fetal and infant development following the WHO guidelines (World Health Organization, 2007).

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Table 1. Descriptive data of the sample

		Mean (SD)* % (n)#
INFANT VARIABLES		
Gender	Boys	51.9 (82)#
	Girls	48.1 (76)#
Breastfeeding (months)		7.78 (4.1)*
OBSTETRIC VARIABLES		
Parity	Primiparous	48.2 (66)#
	Multiparous	52.8 (92)#
Birth weight (grams)		3,297.01 (427.1)*
Gestational age (weeks)		39.52 (1.23)*
SMOKING VARIABLES		
Prenatal	Non-smoking	62.7 (99)#
	SHS exposure	17.1 (27)#
	Smoking	20.3 (32)#
Postnatal	No exposure	65.2 (103)#
	SHS exposure	34.8 (55)#
SOCIODEMOGRAPHIC VARIABLES		
Mother's age (years)		31.42 (4.4)*
Socioeconomic Status	Low	8.2 (13)#
	Medium	52.5 (83)#
	High	39.2 (62)#
PARENT-INFANT PSYCHOLOGICAL VARIABLES		
Prenatal anxiety (total score)		17.2 (8.8)*
General psychopathology (total score)	6 months[b]	18.7 (10.9)*
	30 months[a]	14.8 (6.9)*
	30 months[b]	18.5 (9.9)*
Attachment (score)	6 months	26.1 (3.5)*
	12 months	25.6 (3.9)*
	30 months	12.6 (6.5)*

[a] Father's responses
[b] Mother's responses

Table 2. Mean scores and Standard Deviations of the cognitive development test

		Mean (SD) Median (IQR)*
6 months		
BSID-II	Mental development index	94.08 (10.37)
	Psychomotor development index	85.37 (14.32)
	Cognitive development age	5.60 (0.89)
	Social development age	5.60 (0.82)
	Language development age	5.17 (1.2)
	Motor development age	4.78 (0.67)
12 months		
BSID-II	Mental development index	98.72 (11.38)
	Psychomotor development index	90.08 (12.44)
	Cognitive development age	10.30 (0.94)
	Social development age	9.85 (1.48)
	Language development age	9.99 (1.03)
	Motor development age	9.63 (1.16)
MBCDI	Prelinguistic vocalizations	70.53 (29.11)
	Early comprehension	87.19 (11.64)
	Sentence comprehension	60.63 (25.34)
	Early language production (Imitation)	35.23 (10.24)*
	Early language production (Question)	21.50 (9.50)*
	Early language production (Name)	19.06 (9.67)*
	Word comprehension	53.09 (26.51)
	Word production	57.52 (29.28)
	Nonverbal language	58.62 (29.08)
30 months		
BSID-II	Mental development index	100.82 (3.86)
	Psychomotor development index	98.50 (3.70)
	Cognitive development age	32.93 (5.46)*
	Social development age	32.34 (5.19)*
	Language development age	33.53 (5.84)*
	Motor development age	33.65 (6.27)*
PPVT-III	Language IQ	110.32 (11.18)

BSID-II: Bayley Scales for Infant Development (Second Edition)
MCCDI: MacArthur-Bates Communicative Development Inventories
PPVT-III: Peabody Picture Vocabulary Test-Third Edition

Table 3. Sample characteristics according to MSDP

		NON-SMOKING (n=99)	SHS EXPOSURE (n=27)	SMOKING (n=32)	
		Mean (SD)* % (n)#	Mean (SD)* % (n)#	Mean (SD)* % (n)#	F (p)* Chi (p)#
OBSTETRIC VARIABLES					
Childbirth	Difficult	48.5 (47) #	31.0 (9) #	37.5 (12) #	2.372 (0.305)#
	Not difficult	51.5 (50) #	69.0 (20) #	62.5 (20) #	
Parity	Primiparous	60.8 (59) #	50.0 (15) #	58.0 (18) #	0.815 (0.665)#
	Multiparous	39.2 (38) #	50.0 (15) #	42.0 (13) #	
Birth weight (grams)		3316.48 (416.43)*	3154.07 (406.36)*	3197.01 (427.20)*	1.957 (0.145)*
Gestational age (weeks)		39.57 (1.13)*	39.15 (1.62)*	39.23 (1.15)*	1.585 (0.208)*
INFANT VARIABLES					
Breastfeed (months)		5.57 (5.16)*	4.00 (4.24)*	5.09 (4.99)*	1.062 (0.348)*
Gender	Boy	47.5 (47)#	55.56 (15) #	62.5 (20) #	2.361 (0.307)#
	Girl	52.5 (52) #	44.4 (12) #	37.5 (12) #	
SOCIODEMOGRAPHIC VARIABLES					
Mother's age (years)		32.13 (3.80)*	29.48 (5.43)*	30.88 (4.80)*	4.311 (0.015)*
Scocioeconomic status	Low	30.77 (4) #	38.46 (5) #	30.77 (4) #	
	Medium	56.62 (47) #	21.69 (18) #	21.69 (18) #	14.670 (0.005)#
	High	77.42 (48) #	6.45 (4) #	16.13 (10) #	
PARENT-INFANT PSYCHOLOGICAL VARIABLES					
Prenatal anxiety (total score)		15.90 (7.90)*	19.95 (11.12)*	18.54 (8.64)*	2.189 (0.087)*
General	6 months[b]	16.43 (9.32)*	24.42 (11.80)*	21.87 (13.15)*	3.573 (0.033)*
Psychopathology	30 months[a]	14.55 (6.09)*	13.13 (5.79)*	16.73 (9.54)*	0.828 (0.442)*
(total score)	30 months[b]	17.61 (10.24)*	20.56 (12.25)*	19.81 (8.13)*	0.494 (0.613)*
Attachment (score)	6 months	25.49 (3.94)*	26.89 (2.49)*	27.20 (2.62)*	2.054 (0.087)*
	12 months	25.30 (4.64)*	26.07 (2.46)*	26.73 (1.61)*	0.662 (0.519)*
	30 months	13.03 (7.377)*	10.77 (2.891)*	12.78 (5.725)*	0.627 (0.536)*

[a] Father's responses

[b] Mother's responses

Table 4. Adjusted development scores according to smoking groups

		Non-smoking[a] (99)	**SHS exposure[b] (27)**	**Smoking[c] (32)**		**Post-hoc analysis Tukey Kruskal-Wallis***		
		Mean (SD) Median (IQR)*	Mean (SD) Median (IQR)*	Mean (SD) Median (IQR)*	F (p) Chi (p)*	a-b	a-c	b-c
6 months								
Adjusted BSID-II scores	Mental development index	94.21 (3.14)	92.94 (3.83)	94.31 (4.14)	1.640 (0.197)	0.197	0.992	0.278
	Psychomotor development index	89.06 (5.17)	85.09 (7.35)	87.02 (6.12)	1.601 (0.208)	0.981	0.181	0.440
	Cognitive development age	5.60 (0.34)	5.56 (0.35)	5.61 (0.35)	0.224 (0.799)	0.848	0.966	0.793
	Social development age	5.61 (0.34)	5.54 (0.33)	5.62 (0.27)	0.513 (0.600)	0.603	0.995	0.659
	Language development age	**5.40 (0.46)**	**5.01 (0.41)**	**4.92 (0.43)**	**9.692 (0.001)**	**0.010**	**0.001**	0.862
	Motor development age	4.85 (0.22)	4.79 (0.22)	4.70 (0.21)	4.027 (0.020)	0.134	0.021	0.946
12 months								
Adjusted BSID-II scores	Mental development index	98.49 (3.80)	97.90 (2.32)	99.67 (3.27)	2.918 (0.061)[1]	0.714	0.224	0.131
	Psychomotor development index	**91.41 (2.88)**	90.90 (2.55)	**89.32 (2.66)**	**4.797 (0.010)**	0.158	**0.014**	0.777
	Cognitive development age	10.29 (0.37)	10.24 (0.36)	10.32 (0.27)	0.490 (0.614)	0.742	0.883	0.589
	Social development age	9.78 (0.55)	9.89 (0.44)	9.95 (0.46)	1.480 (0.231)	0.587	0.250	0.906
	Language development age	**10.22 (0.31)**	**9.87 (0.28)**	9.95 (0.46)	**5.142 (0.007)**	**0.009**	0.470	0.589
	Motor development age[1]	9.59 (0.46)	9.62 (0.44)	9.66 (0.27)	0.360 (0.698)	0.938	0.682	0.929
Adjusted MBCDI scores	**Prelinguistic vocalizations**	**71.34 (6.32)**	**67.65 (5.98)**	70.28 (6.50)	**3.666 (0.020)**	**0.019**	0.686	0.250
	Early comprehension	**89.93 (2.53)**	**85.78 (2.45)**	**86.17 (2.55)**	**11.148 (0.001)**	**0.001**	**0.002**	0.828
	Sentence comprehension	**61.21 (3.36)**	59.66 (2.54)	**58.54 (3.44)**	**4.581 (0.012)**	0.074	**0.017**	0.990
	Early language production (Imitation)	35.08 (4.14)*	35.11 (4.70)*	35.37 (1.19)*	3.218 (0.200)*	0.645*	0.076*	0.280*
	Early language production (Question)	21.33 (3.52)*	21.35 (4.73)*	21.46 (1.92)*	1.053 (0.591)*	0.796*	0.383*	0.323*
	Early language production (Name)	18.97 (3.94)*	18.91 (4.85)*	19.10 (1.18)*	1.563 (0.458)*	0.879*	0.270*	0.229*
	Word comprehension[1]	**54.35 (3.90)**	**51.02(4.52)**	**51.59 (4.95)**	**8.662 (0.001)**	**0.001**	**0.005**	0.866
	Word production	57.97 (7.36)	56.79 (8.70)	57.52 (7.70)	0.447 (0.640)	0.763	.717	0.997
	Nonverbal language	58.91 (6.92)	58.42 (6.07)	58.60 (8.47)	0.061 (0.941)	0.946	0.976	0.995
30 months								
Adjusted BSID-II scores	**Mental development index**	**100.82 (3.86)**	**97.08 (3.57)**	**98.15 (3.73)**	**7.156 (0.001)**	**0.004**	**0.020**	0.805
	Psychomotor development index	**98.50 (3.70)**	**95.45 (4.35)**	**98.21 (3.94)**	**6.729 (0.002)**	**0.001**	0.930	**0.019**
	Cognitive development age	32.39 (2.39)*	32.72 (1.77)*	33.02 (2.75)*	3.926 (0.140)*	0.387*	0.057*	0.361*
	Social development age	32.06 (2.21)*	31.53 (1.65)*	31.88 (2.20)*	3.599 (0.165)*	0.057*	0.510*	0.330*
	Language development age	**35.00 (1.98)***	**33.76 (1.68)***	33.12 (2.36)*	**13.250 (0.001)***	**0.010***	**0.002***	0.523*
	Motor development age	33.25 (2.37)*	32.71 (2.10)*	33.59 (3.93)*	3.922 (0.141)*	0.260*	0.182*	0.051*
Adjusted PPVT-III scores	Language IQ	110.10 (2.97)	110.65 (1.45)	110.23 (1.81)	0.375 (0.688)	0.586	0.966	0.806

Each score was adjusted by child gender, maternal prenatal anxiety, birth weight, gestational age at birth, parity, SES, infant age at cognitive assessment, infant postnatal smoke exposure, months of breastfeeding, mother-infant attachment, and parent psychopathological and health general status. With the adjusted variables, we used parametric (ANOVA) and non-parametric (Mann-Whitney) test.

[1] The Welch statistics have been applied as group variance is not homogeneous.

* Scores not normally distributed.

Significance level=0.02.

BSID-II: Bayley Scales for Infant Development (Second Edition); MCCDI: MacArthur-Bates Communicative Development Inventories; PPVT-III: Peabody Picture Vocabulary Test-Third Edition