

# Pilot and Feasibility Studies

## Do fetuses move their lips to the sound that they hear? An observational feasibility study on auditory stimulation in the womb

--Manuscript Draft--

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<b>Abstract:</b>	<p><b>Background:</b> We investigate in this feasibility study whether specific lip movements increase prenatally when hearing a particular sound. We hypothesised that fetuses would produce more mouth movements resembling those required to make the sound stimulus they heard (i.e. mouth stretch) compared with a no-sound control group who heard no specific auditory stimuli. Secondly, we predicted that fetuses hearing the sound would produce a similar number of mouth movements unrelated to the sound heard (ie lip pucker) as the no-sound group of fetuses.</p> <p><b>Methods:</b> In an observational feasibility study, 17 fetuses were scanned twice at 32 and 36 weeks gestation, and two different types of mouth movements recorded. Three fetuses received an auditory stimulus, and 14 did not. A generalised mixed effects log-linear model was used to determine statistical significance.</p> <p><b>Results:</b> Fetuses in the sound group performed one specific mouth movement (mouth stretch) significantly more frequently than fetuses in the no-sound group. A significant interaction between group and gestational age indicates that there was differential change in this specific movement as age increases (<math>X^2 = 7.58</math> on 1 df, <math>p = .006</math>), with the no sound group showing a decline of 76% between 32 weeks and 36 weeks (<math>p &lt; .001</math>), whereas the sound group showed no significant change over time (<math>p = 0.41</math>). There was no significant difference between the sound group and no sound group in the frequency of lip puckering - the second, unrelated mouth movement (<math>p = 0.35</math>).</p> <p><b>Conclusions:</b> These results suggest that a sound stimulus is associated with an increase in specific, rather than general, mouth movements. The results are informative for the development of infant speech, and potentially could also lead to a diagnostic test for deafness in utero. More research is needed to replicate this research with a randomised design and with a range of different auditory stimuli which would be produced with different mouth movements, such as "o" which would be seen as pursed lips.</p>	
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auditory stimulation in the womb.**

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**Running title:** Auditory stimulation in the womb

# Abstract

**Background:** We investigate in this feasibility study whether specific lip movements increase prenatally when hearing a particular sound. We hypothesised that fetuses would produce more mouth movements resembling those required to make the sound stimulus they heard (i.e. mouth stretch) compared with a no-sound control group who heard no specific auditory stimuli. Secondly, we predicted that fetuses hearing the sound would produce a similar number of mouth movements unrelated to the sound heard (ie lip pucker) as the no-sound group of fetuses.

**Methods:** In an observational feasibility study, 17 fetuses were scanned twice at 32 and 36 weeks gestation, and two different types of mouth movements recorded. Three fetuses received an auditory stimulus, and 14 did not. A generalised mixed effects log-linear model was used to determine statistical significance.

**Results:** Fetuses in the sound group performed one specific mouth movement (mouth stretch) significantly more frequently than fetuses in the no-sound group. A significant interaction between group and gestational age indicates that there was differential change in this specific movement as age increases ( $\chi^2 = 7.58$  on 1 df,  $p = .006$ ), with the no sound group showing a decline of 76% between 32 weeks and 36 weeks ( $p < .001$ ), whereas the sound group showed no significant change over time ( $p = 0.41$ ). There was no significant difference between the sound group and no sound group in the frequency of lip puckering - the second, unrelated mouth movement ( $p = 0.35$ ).

**Conclusions:** These results suggest that a sound stimulus is associated with an increase in specific, rather than general, mouth movements. The results are informative for the development of infant speech, and potentially could also lead to a diagnostic test for deafness in utero. More research is needed to replicate this research with a randomised design and with a range of different auditory stimuli which would be produced with different mouth movements, such as "o" which would be seen as pursed lips.

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**Keywords:** prenatal stimulation; fetal hearing; fetal mouth movement

# Background

From birth, infants produce silent movements resembling the lip movements necessary for speech.

A seminal study reported by Trevarthen [1] in which one 7 week old girl silently produced lip and mouth movements which resembled the mouth movements of a female speaker reading a word list, has been interpreted as the basis of the intention to speak. These mouth movements indicated that very young infants when hearing language in their first weeks after birth produce mouth and lip movements similar to those necessary to replicate speech. In addition, infants just after birth have been shown to imitate silently mouth movements required to produce language sounds, even when no sound was produced [2].

One way in which this ability could develop prenatally was proposed by Green and Wilson [3]. They argued that randomly produced lip and jaw movements during fetal development could conceivably create sensorimotor pathways that could serve as precursors of early speech. They based their conjecture on the well-established suggestion that neuronal firing resulting in leg and hand movements are precursors of walking and grasping movements (e.g. [4;5]). Neuronal activation elicits early limb movements and these limb movements in turn help to consolidate the pathways which shape purposeful movements such as reaching or walking (e.g. [4;5]). Hence, early activation of specific mouth and jaw movements such as a jaw drop with elongation of lips in the vertical axis could be a precursor necessary for speech sounds such as “a” and pursing lips could be a precursor to producing the lip movement necessary for the sound “o”.

Prenatal cognitive development has been tested in relation to sound and light stimulation. For example, Horimoto et al. [6] reported that fetuses between 32 and 34 weeks gestation showed a high incidence of mouth movements that were later during gestation correlated with non-rapid eye movements. However, the mouth movements which have been reported in previous studies are general movements that do not have specific definitions, such as a smile or grimace.

1 The following sequential order of development has been found for specific fetal oral movements,  
2 namely jaw opening, jaw closing, tongue movement, and lip movement [7], whereby spontaneous  
3 movements of the jaw appear as early as 11 weeks during prenatal development [8]. Regarding first  
4 vocalizations in infancy, jaw opening and closing are primary movements during babbling [9] which  
5 can be distinguished from other mouth openings such as smiling [10]. Green, Moore and Reilly [11]  
6 investigated the sequential development of jaw and lip control and found in an analysis of children  
7 aged 1 to 6 years of age developed sequentially, with jaw movements preceding lip movements.  
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19 By the second trimester, the fetal auditory cortex is reorganized by external stimulation (e.g.  
20 [12;13]). In the study by Kisilevsky et al., [13] which examined maturation of fetal responding to  
21 airborne auditory stimuli, differential responding occurred as a result of fetal maturation during the  
22 third trimester. Hence, we selected two gestational ages in the third trimester to examine whether  
23 we might find maturational changes in reaction to an auditory stimulus. If fetuses were exposed to  
24 auditory rhythmic stimulation [14], then this might be reflected in their production of movement  
25 patterns, specifically movement patterns of the jaw and lips. Given that fetal mouth movements  
26 develop but have to date not been analysed to the level of specificity required, it is essential to  
27 establish whether there is a relationship between mouth movements and sound stimulation. If jaw  
28 and lip movements are produced prenatally in response to sound before the ability to produce  
29 speech develops, then this would support the argument that precursors of language are rooted in  
30 fetal development. In particular, we argue that if types of jaw and lip movements vary between  
31 fetuses who do not hear any sounds during scans and fetuses who hear specific sounds, there might  
32 be reason to believe that lip and jaw movements are pre-cursors of silent pre-speech movements  
33 which can be observed in neonates.  
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57 The production of the auditory stimulus presented in this study involved predominantly jaw  
58 movements, allowing us to determine whether the response to this stimulus was specific (i.e., only  
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1 jaw movements were produced) or general (i.e., jaw and lip movements were produced). In order to  
2 investigate this question we studied two groups of fetuses: one that was presented with the  
3 auditory stimulus during scanning and a second no-sound control group of fetuses that was not  
4 presented with any sound stimulus. We expected that those fetuses presented with the specific  
5 sound would produce mouth movements consistent with the sound when contrasted with another  
6 mouth movement that typically manifests with the same frequency in normally developing fetuses.  
7 This feasibility study was designed to establish whether it is possible to test fetal fine grained  
8 reactions to specific sound stimulation.  
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## 21 **Methods**

### 22 **Ethics**

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28 Ethical permission for the feasibility study was obtained from the County Durham and Tees Valley 2  
29 Research Ethics Committee (REC Ref: 08/H0908/31 and County Durham and Tees Valley 2 Research  
30 Ethics Committee REC Ref: 11/NE/03/61) and the research and development department of James  
31 Cook University Hospital, as well as the Durham University (Department of Psychology ethics  
32 committee). All mothers gave informed written consent.  
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### 43 **Stimulus**

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46 The auditory stimulus consisted of multiple presentations of the sound MA (/ma:/ in the  
47 International Phonetic Alphabet). The MA sound was spoken in a female voice, was 0.40s in length,  
48 and was repeated 8 times with 0.80s of silence between each presentation. This cluster of eight MA  
49 sounds was then repeated for the duration of the scan, with 6.0s of silence between each cluster.  
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55 The auditory stimulus was played on a Sandisk Sansa Clip portable MP3 player, attached to a  
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[15], who report research showing that the speaker should not be placed on the mother's abdomen, for this study the speaker was held at a distance of 3cm above the mother's abdomen near the ear of the fetus for the duration of the stimulus presentation. The sound pressure level at a distance of 3cm from the speaker was 94dB, measured with a Precision Gold N05CC Digital Sound Meter (with a measurement range of 30 - 100dB with an accuracy of  $\pm 1.5$ dB), although uterine attenuation will have reduced the sound level for the fetus by approximately 20 - 35dB [16;17]. The auditory stimulus contained frequencies between 0Hz and 11kHz, with most output in the 0.6 - 1.6kHz and 2.6 - 3.6kHz regions. These frequencies are audible to fetuses from 29 weeks gestational age [16]. Although, in a study of newborn auditory matching, Chen, Striano and Rakoczy [18] only measured responding to an auditory stimulus during its presentation, we measured responding during the presentation of sound and during the intervening seconds of silence. The inclusion of short periods of silence following the presentation of sound was deemed appropriate as fetuses may be slower to respond to sound stimuli than newborns due to being less developmentally mature. The short periods of silence allow the fetuses time to respond to the recently presented auditory stimuli. The relative frequencies of jaw and lip movements during the presentation of the auditory stimuli were analysed and compared with the no-sound control group who did not experience the auditory stimulus.

## Participants

Mothers who had completed their normal 20-week anomaly scans were invited to participate in this study. All fetuses participating in this study were completely healthy as determined by their 20 week scan. A convenience sample of seventeen mothers was recruited for this feasibility study; three mothers whose fetuses were exposed to the auditory stimulus (1 boy and 2 girls), the sound group and 14 mothers whose fetuses were not exposed to any stimulus (7 boys and 7 girls), the no-sound group. The no-sound group participants were recruited through the midwives of the antenatal unit of the James Cook University Hospital, Middlesbrough, UK, and sound group participants through the Hypnobirthing group in London at the Harley Street Ultrasound Centre

1 (www.thewisehippo.com) following approved ethical procedures. During consent and before each  
2 procedure, mothers were made aware that the scans were for research purposes and were not  
3 routine medical scans. Given that maternal stress, attachment, anxiety, and depression are known to  
4 affect fetal behavior (e.g., [19, 20, 21, 22]), the two groups were assessed for these factors with the  
5 Perceived Stress Scale (PSS [23]), Antenatal Maternal Attachment Scale (AMAS [24]), and the  
6 Hospital Anxiety and Depression Scale (HADS [25]).  
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## 18 Procedure

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21 All participating mothers received scans at 32 and 36 weeks gestational age, with fetuses being  
22 scanned for approximately 900-1200 seconds, with the maximum time of the scan determined by  
23 the British Medical Ultrasound Society (BMUS) guidelines. The scan times of 32 and 36 weeks were  
24 chosen as Kisilevsky et al [13] identified that it was more likely to get reactions to sound in the latest  
25 stages of pregnancy. The scans took place either in the radiography department of James Cook  
26 University Hospital, where mothers had previously undergone their routine 12 and 20 week medical  
27 scans, or in the London Ultrasound centre. The scanning took place with mothers lying in a darkened  
28 room on their back or on their side, depending on the position of the fetus and how comfortable  
29 mothers were. The fetal face and upper torso were visualized both by means of 4-D color full frontal  
30 or facial profile ultrasound recordings, as well as sequences of traditional 2-D monochrome images.  
31 The scans were recorded for off line analysis with a GE Voluson E8 Expert Ultrasound System using a  
32 GE RAB4–8L Macro 4D Convex Array Transducer. Mothers were provided with a DVD copy of their  
33 scans.  
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## 54 Measures

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57 Scan recordings were used to code mouth movements using the Fetal Observable Movement System  
58 (FOMS) [26], an adaptation of the Facial Action Coding System [27], which has been found to be  
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1 reliable in previous research [28;29]. Following established procedures [29] two types of mouth  
2 movements were identified for analysis: *mouth stretch* and *lip pucker*.  
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5  
6 Mouth stretch is defined by the lower jaw being pulled down by the action of the external pterygoids  
7 and digastricus muscles, so that the mouth is actively opened. The opening is stretched such that  
8  
9 the longest axis is the vertical plane. The cheeks are stretched and flattened and the skin on the chin  
10  
11 also may become bulged.  
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15  
16 Lip pucker in contrast, is defined by the lips narrowing and pursing with the lips protruding forwards.  
17  
18 This is caused by the incisivii labii superioris and incisivii labii inferioris muscles, which pull the  
19  
20 corners of the lips medially. The lips usually appear as if contracted and the mouth opening will look  
21  
22 smaller and rounded. There also may be some bulging of the chin as the skin of the chin is pulled  
23  
24 upwards towards the lips. In normally developing fetuses, these two mouth movements are  
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26 produced with relatively equal frequencies at the gestational ages scanned in this study [26].  
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34 The aim in this feasibility study was to analyse up to 600 seconds of codable scan for each fetal scan.  
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36 Codable sections of the scan for the control scans were sections where the fetal face was visible, and  
37  
38 where the pocket of amniotic fluid was present to allow a clear image. For the sound group, coding  
39  
40 occurred during the presentation of sound and during the intervening seconds of silence. As fetal  
41  
42 movements differ as a function of the movement state of the fetus [30], it is essential that all of the  
43  
44 fetuses were in an active state during scanning. Assessment of the movement behaviour showed  
45  
46 that all fetuses were in the active states of 2F or 3F during scanning, as assessed by their gross body  
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48 movements and eye movements according to Nijhuis et al.'s four-state categorisation [30], and not in  
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50 states 1F (totally passive) or 4F (overly active).  
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## 59 Statistical analysis 60 61 62 63 64 65

1 Reflecting the longitudinal structure of the measurements and the non-normality of the count  
2 responses, a Poisson log-linear mixed effects analysis [31] was used to assess the effect of  
3  
4 experimental group and gestational age, and the interaction between them using the *glmer* function  
5  
6 of the *lme4* library in R [32]. A mixed effects analysis has been shown to be superior to standard  
7  
8 repeated measures analysis of variance for experimental data [33]. Moreover, imbalance in the  
9  
10 number of participants in each treatment arm, and in the number of scans contributed by each  
11  
12 mother can easily be accounted for. The analysis models the number of *mouth movements* of  
13  
14 different types as a count variable adjusted by the length of analysed scan as an exposure variable,  
15  
16 and a random individual fetus effect. The individual random fetus effect allows for individual  
17  
18 variability between fetuses in their overall propensity to mouth movements and is assumed to be  
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20 normally distributed.  
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28 Formally, we can write the model as  
29

$$30 \quad M_{it} \sim \text{Poisson}(\lambda_{it})$$

31 with  
32

$$33 \quad \log(\lambda_{it}) = \log(\text{scan length}_{it}) + \beta_0 + \beta_1 \text{ gestational age}_{it} + \beta_2 \text{ treatment}_i \\
34 \quad + \beta_3 (\text{gestational age} * \text{treatment})_{it} + u_i \\
35 \quad u_i \sim \text{Normal}(0, \sigma_f^2),$$

36 where  $M_{it}$  are the mouth movement counts for fetus  $i$  at gestational age  $t$ ,  $\lambda_{it}$  is the underlying  
37  
38 Poisson rate,  $\beta_0$  to  $\beta_3$  are unknown regression coefficients and  $\sigma_f^2$  is the individual within-fetus  
39  
40 variance. The indices to the individual covariates show which of them vary over time in our model.  
41  
42 Thus gestational age, is recorded at each scan, whereas the treatment condition (sound/no sound) is  
43  
44 constant for each fetus.  
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53 A test for overdispersion for count data was carried out on the full interaction model using the  
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55 methodology of Mancuso[34]. If the overdispersion test indicated no overdispersion, then  
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1 significance of terms was assessed through analysis of deviance likelihood ratio test, examining  
2 changes of deviance between fitted models, and comparing to a chi-squared distribution with the  
3 appropriate number of degrees of freedom. Bolker et al [35] provides full details of fitting and  
4 testing Poisson log-linear mixed effects for count data.  
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## 10 11 **Results**

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13 Comparing mothers in the two groups we analysed their results on a number of scales. On the  
14 Perceived Stress Scale (PSS [23]), mothers in the sound group were similar to those in the no-sound  
15 group at 32 weeks ( $t(15) = 0.16, p = .87$ ) and at 36 weeks ( $t(15) = 0.18, p = .86$ ). Scores on the  
16 Antenatal Maternal Attachment Scale (AMAS [24]) also did not differ between mothers in the sound  
17 and no-sound control groups either at 32 weeks ( $t(15) = 0.85, p = .41$ ) or 36 weeks ( $t(15) = 0.50, p =$   
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1 movements per hundred seconds of codable scan time. Thus, frequency of movements can be  
2 readily compared between groups with differing scan times in the descriptive Tables 1 and 2.. Below  
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4 we report the results for “mouth stretch” and “lip pucker”.  
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6

## 7 Mouth Stretches

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10 The mouth stretch means from the fetuses at 32 and 36 weeks gestational age can be seen in Table  
11 1 and Fig. 1a. These suggest that fetuses at both gestational ages produce more frequent mouth  
12 stretches in the sound group compared with the no-sound control group. These observations were  
13 confirmed by the main effects Poisson linear mixed model. The overdispersion test gave a  
14 dispersion parameter estimate of 0.89, which is under one, and therefore indicates no  
15 overdispersion. There was a significant main effect of group,  $X^2= 5.78$  on 1 df ,  $p = .01$ , and of  
16 gestational age ( $X^2= 10.78$  on 1 df ,  $p = .001$ ), demonstrating that fetuses in the sound group  
17 performed mouth stretches more frequently than fetuses in the no-sound control group, and there  
18 was a general trend towards fewer mouth movements as gestational age increased. When an  
19 interaction model was fitted, the interaction between group and gestation was also significant ( $X^2=$   
20  $7.58$  on 1 df ,  $p = .006$ ), indicating that there was differential change in mouth stretch rate as  
21 gestational age increases. For the sound group there was no evidence of a change in the rate of  
22 mouth stretch ( $\beta = 0.49$ , 95%  $CI = [-0.71, 1.69]$ ,  $\exp(\beta) = 1.63$  ,  $p = .41$ ); for the no-sound group  
23 there was a decrease of 76% in the rate of mouth stretch ( $\beta = -1.41$ , 95%  $CI = [-2.17, -0.65]$ ,  
24  $\exp(\beta) = 0.24$ ,  $p < .001$ ).  
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48 **Table 1 about here**

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53 **Figure 1 about here**

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58 These data indicate that fetuses presented with the auditory stimulus MA produce more mouth  
59 stretches than fetuses presented with any specific auditory stimulus. However, it is possible that  
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1 the presentation of the auditory stimulus triggered an increase in the number of mouth movements  
2 in general. In order to test this hypothesis, the frequency with which fetuses performed the second  
3 mouth movement, a lip pucker, was established. The lip pucker is a valid mouth movement for  
4 comparison as fetuses at 32 and 36 weeks of age not exposed to any specific stimulation show  
5 similar frequencies of mouth stretches and lip puckers. To corroborate this, we examined  
6 frequencies of lip pucker and mouth stretch in the control group. A paired-samples t-test indicated  
7 that, pooled over the two gestational ages, there was no significant difference between the  
8 frequency of mouth stretches ( $M = .163$  per minute) and lip puckers ( $M = .218$  per minute),  $t(13) =$   
9  $0.63, p = .54$ .

## 23 Lip Pucker

24 The lip pucker mean counts from the fetuses at 32 and 36 weeks gestational age can be seen in  
25 Table 2 and Fig. 1b showing only small differences in the frequencies with which fetuses pucker their  
26 lips depending on gestational age and group. Across both gestational ages, it appears that a similar  
27 amount of lip puckers were produced by the control group and the sound group. This was tested  
28 using the mixed effects Poisson model as before. Again, the overdispersion test indicated that no  
29 overdispersion was present in the data, with the dispersion parameter of 0.516 being less than one.  
30 Results indicate no statistically significant main effect of group,  $X^2 = 0.87$  on 1 df,  $p = .35$ , no  
31 significant main effect of gestational age,  $X^2 = 0.01$  on 1 df,  $p = .93$ , and no interaction between  
32 group and gestation,  $X^2 = 1.64$  on 1 df,  $p = .20$ .

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51 **Table 2 about here**  
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54 These data suggest that the frequency with which fetuses show lip pucker lip movements are  
55 independent of the presentation of an auditory stimulus. Additionally, there is no evidence that  
56 gestational age affects the rate of lip puckers. From this result we can infer that the presentation of  
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1 the auditory stimulus MA does not cause an increase in the frequency of mouth movements in  
2 general. Rather, it increases the frequency of a specific mouth movement corresponding to the MA  
3 sound, with a jaw drop and the mouth stretching in the vertical plane.  
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## 9 **Discussion**

10 Results of this study indicate that fetuses respond to a specific sound MA with a specific mouth  
11 movement which mimics the sound heard, namely a mouth stretch which involves a jaw drop. There  
12 are a number of researchers who argue that given the precocity of the functional development of  
13 the auditory system, the abilities shown in new-born babies must have their origin in prenatal life  
14 [36;13]. Given that research [37] has established that the fundamental frequency (F0) of vowels are  
15 well transmitted to the fetus, using the sound MA seemed to be an ideal candidate in this study.  
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18 However, in order to eliminate the possibility that fetuses would respond with more mouth  
19 movements in general we analysed the occurrence of another type of mouth movement, namely  
20 pursing of lips, which in the control no-sound group occurred with similar frequencies to mouth  
21 stretching. We found that upon hearing MA, fetuses did not respond with an increase in pursing of  
22 their lips. Given the results by Green et al. [11] who found that control over vertical movements of  
23 lips and jaw during speech developed sequentially with jaw movements preceding lip movements,  
24 this might explain why we found a relationship between hearing the MA sound and producing the  
25 mouth stretch. Ferronato, Domellöf and Rönqvist [38] suggest that postnatally speech stimuli are  
26 special stimuli which elicit specific behavioural reactions. They argue that the pairing of certain  
27 acoustic stimuli with defined motor activities (e.g., rhythmic sounds with rhythmic movements)  
28 demonstrate that the “human brain is primed with the body” (p. 3). This according to Ferronato et  
29 al. [38] could indicate that auditory input and behavioural output might play a role in the integration  
30 between external and internal information which is essential for learning in general and language  
31 acquisition in particular. In terms of differential responding to sounds over the duration of the third  
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1 trimester, fetuses did not show a significant increase in the production of mouth movements as a  
2 consequence of maturation. However, there is a numerical increase in mean responses as  
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4 gestational age increases, which needs to be further investigated.  
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9 Turning to the observed changes over time, we identified a significant interaction between the  
10 sound and no-sound groups in the slope of the rate of mouth stretch in response to the stimulus of  
11 “MA”. The no sound group showed declining rate of mouth stretch, whereas the sound group  
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13 showed no such decline. Fetal movements in general tend to decrease with gestational age. Our  
14 results provide evidence that appropriate mouth movements associated with specific stimuli do not  
15 decrease in this way.  
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24 This feasibility study was not a randomised control trial but used a convenience sample. However we  
25 consider it unlikely that fetuses of mothers who did not consent to participate were different to  
26 those who participated in our study, and so there was minimal selection bias.  
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## 32 33 34 **Conclusion** 35 36

37 This feasibility study established that it is possible to test specific fetal fine grained reactions to  
38 sound stimulation. More research is needed to develop this feasibility study. Firstly a randomised  
39 controlled trial is needed with balanced numbers of treatment and control participants. Additionally,  
40 a range of different auditory stimuli should be examined to determine whether the fetus is reacting  
41 to a specific MA sound, or to any general auditory stimulus such as white noise. Additionally, a  
42 greater range of specific sounds could also be examined in order to fully explore how the fetus  
43 produces not only random mouth movements but specific pre-speech movements.  
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56 The potential implications of this work are twofold. Firstly, this work is likely to provide knowledge  
57 on the relationship of prenatal reaction to specific sounds and postnatal language development.  
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Secondly, there is a possibility that lack of reaction of the fetus to specific sounds could be used as a pre-natal diagnostic test for deafness.

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## Authors' contributions

NR and VR conceived the study. NR and BF wrote the paper. The coding of the fetal scans was carried out by LB and JA. BF also carried out the statistical analysis . All authors read and approved the final manuscript.

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## Declarations

'The authors declare that they have no competing interests'

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**Table Legends**

**Table 1. Average number of mouth stretches per scan and rates of mouth stretching per 100 second of scan [with 95% confidence intervals] for the sound and control groups by gestational age**

**Table 2. Average number of lip puckers per scan and rates of lip puckering per 100 second of scan [with 95% confidence intervals] for the sound and control groups by gestational age**

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## Figure legends

**Fig. 1.** The effect of gestational age and presentation of an auditory stimulus on the frequency of (a) mouth stretches and (b) lip puckers. Inset images provide examples of (a) mouth stretch and (b) lip pucker movements.

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**Table 1. Average number of mouth stretches per scan and rates of mouth stretching per 100 second of scan [with 95% confidence intervals] for the sound and control groups by gestational age**

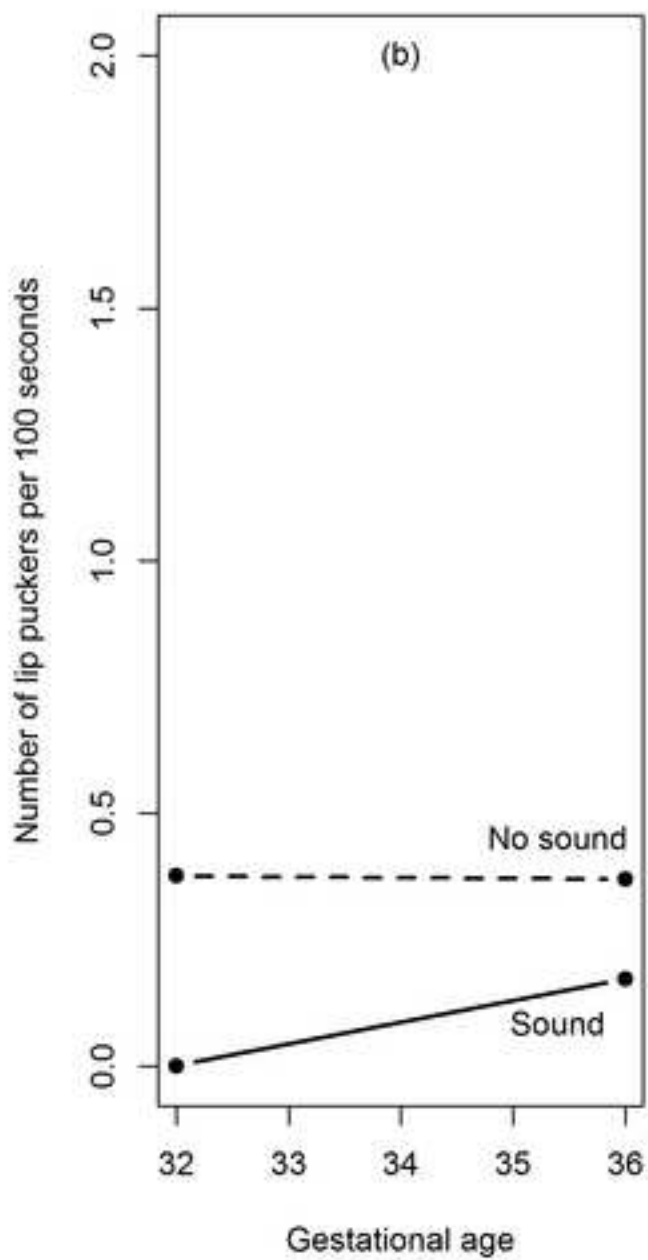
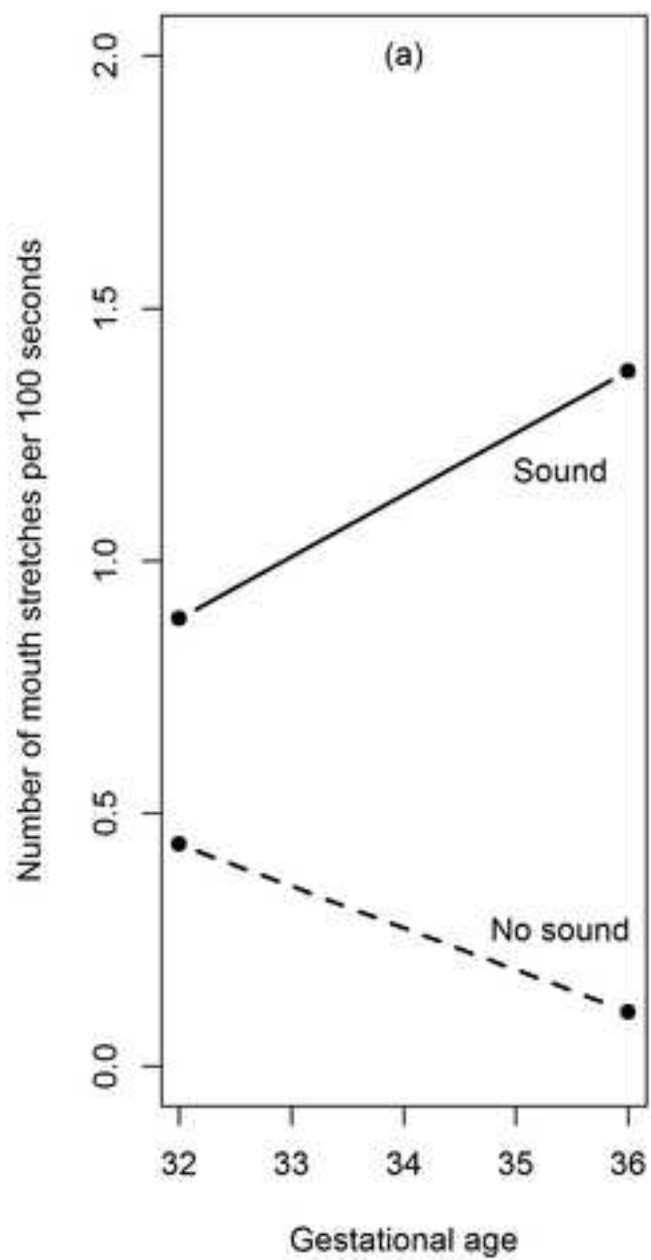
	Average Number of Movements Observed per Scan		Relative Frequency (movements per 100 seconds of scan)	
	32 weeks	36 weeks	32 weeks	36 weeks
<b>Sound</b>	1.67 [0.63, 3.65]	2.67 [1.26, 5.23]	0.89 [0.33, 1.94]	1.38 [0.65, 2.60]
<b>Control</b>	2.50 [1.77, 3.44]	0.64 [0.32, 1.17]	0.44 [0.31,0.60]	0.11 [0.15,0.20]

Note: 95% confidence intervals calculated using Byar's method for rates [39]

**Table 2. Average number of lip puckers per scan and rates of lip puckering per 100 second of scan [ with 95% confidence intervals] for the sound and control groups by gestational age**

	Average Number of Movements Observed per Scan		Relative Frequency (movements per 100 seconds of scan)	
	32 weeks	36 weeks	32 weeks	36 weeks
<b>Sound</b>	0 [0.00, 0.82]	0.33 [0.03, 1.55]	0 [0.00, 0.43]	0.17 [0.02, 0.80]
<b>Control</b>	2.14 [1.47, 3.01]	2.21 [1.53, 3.10]	0.38 [0.26, 0.53]	0.37 [0.26, 0.52]

Note: 95% confidence intervals calculated using Byar's method for rates [39]





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