

# Uncultured bacteria found in amniotic fluids of women who experience preterm births

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Researchers from Case Western Reserve University and Yale University have made a significant advancement in understanding the cause behind why some pregnant women suffer from inflammations in the inner womb without any signs of an infection.

Using gene-cloning techniques, researchers discovered that approximately 60 percent of the bacteria present in women with intra-amniotic inflammations were missed by traditional culture testing — considered the gold standard for finding bacterial infections.

The findings were reported in the January issue of the *Journal of Clinical Microbiology*.

For the first time, the researchers identified a comprehensive list of microbial species in intra-amniotic infections by using new DNA methods to track down the presence of bacteria. To increase their accuracy the investigators used a combination of analyses including proteomics results of amniotic fluid and histological analysis of the placenta to corroborate the infection and inflammation.

Intra-amniotic inflammation is known to set off spontaneous births of preterm babies at less than 32 weeks, said Yiping Han, associate professor of dental medicine at Case Western Reserve University and lead investigator on the study of 46 women of which 44 experienced preterm births. Han has previously done a number of research projects examining the link between oral bacteria and preterm birth.

In the present study, bacteria levels from the amniotic fluids of pregnant women with signs or symptoms of preterm births were compared to those from a control group of 16 women without such manifestations and who delivered at term. The amniotic fluid of the control group came from amniocenteses for genetic screenings or analyses to check fetal lung maturity and showed no signs of bacteria even by DNA methods.

"Because culturing is not finding all bacteria present in the amniotic fluid, this calls for new detection methods," Han said. "It is also important to identify which germ is causing the infection and inflammation leading to preterm birth so that antibiotics are initiated early in this pathophysiological chain of events."

In addition to the bacteria identified by cultures by using a new detection process which amplifies the 16SrRNA bacterial gene and clones it in order to identify its sequence, the researchers were able to identify a number of harmful bacteria not detected by cultures of which some have not been previously linked with preterm birth.

"By employing 16s rRNA gene-based polymerase chain reactions (PCR) followed by the clone analysis, we stand to determine the identity and true relationship between intra-amniotic bacterial infection and the onset of preterm birth," said Han.

The researchers also found it was not just one bacterial species causing the inflammation but an abundance of different species in the sample. "Unrecognized, uncultivated or difficult-to-cultivate species may play a key role in initiating spontaneous preterm births," said Han.

These bacteria either reach the placenta through the genital tract or through the blood to the placenta. Han suspects some originate in the mouth which has hundreds of dozens of bacteria present. Among the oral bacteria is *Fusobacterium nucleatum*, which is ubiquitous in the

mouth. Once it enters the blood stream, however, it has been linked to a number of health issues.

Source: Case Western Reserve University

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