Genetics inMedicine | CORRESPONDENCE



# Response to Toutain et al.

We thank Dr. Toutain and colleagues for their correspondence[1]. They point out that their previous work had identified confined placental mosaicism type 3 (CPM3; presence of an abnormal cell line in both cytotrophoblasts and mesenchyme) as a risk factor for birthweight <3rd percentile.[2,3] CPM2 (abnormality confined to mesenchyme) was not a risk factor for low birthweight.

We have reviewed our data to look for this association.[4] For all referrals to the TOMA lab, we found 41/57,539 (0.07%) CPM3 cases that involved a rare autosomal trisomy (RAT), excluding those involving trisomy 16. Such cases were therefore rare in our chorionic villus sampling (CVS) patient referrals (approximately 1 in 1400 cases). For CPM2, RATs, excluding trisomy 16, we observed 191/57,539 (0.33%, approximately 1 in 300 cases).

In our study on outcomes of women with CPM,[5] we had only nine cases of CPM3 involving a RAT other than trisomy 16. Supplemental Table 1 summarizes the trisomic chromosomes involved, reasons for referral, and pregnancy outcomes. The frequency of cases with birthweight <3rd percentile (2/9, 22.2%) was significantly higher than the frequency (13/401, 3.2%) seen in our control group ($P = 0.039$, odds ratio [OR] 8.5, 95% confidence interval [CI] 1.6; 45.1). For CPM2, there were 40 cases in the study with birthweight information available in 37. There were 0/37 with birthweight <3rd percentile, which was not significantly different from the controls (approximate $P = 0.4$, OR 1.34, 95% CI 0.2; 10.4).

Although based on small samples of CPM3 and CPM2 cases, our observations are consistent with the conclusion of Toutain et al. that CPM3, but not CPM2, is associated with increased risk for birthweight <3rd percentile. For CPM3, there was insufficient data to evaluate the risk for other pregnancy complications and for CPM2 we did not note any other significant associations.

There are a number of important differences between our study[5] and those of Toutain et al.[2,3] We have restricted outcome analyses to RATs, i.e., we did not include trisomies 21, 18, 13, or sex chromosome aneuploidy. There also appear to be differences in the frequency of CPM3. For example, in our population the overall frequency of CPM3 involving any type of aneuploidy except trisomy 16 (i.e., RATs, common trisomies, sex chromosome aneuploidies, multiple trisomies, and autosomal monosomy) was 83/57539 (0.14%), while Toutain et al. reported a rate of 23/5512 (0.42%) in their second cohort ($P < 0.0001$, OR 2.9, 95% CI 1.8; 4.6).[3] These differences may be explained by referral for testing of pregnancies with fetal growth restriction (FGR) that had already been detected by ultrasound, differences in maternal age that will affect the frequency of cases with a meiotic origin for the trisomy, and the use of prenatal serum screening that includes pregnancy-associated plasma protein A where very low levels are known to be associated with some RATs and FGR.[6–8] These factors are expected to affect both the frequency of CPM3 and the spectrum of specific trisomies present in the CPM3 group.

We agree with Toutain et al. that when CPM is serendipitously encountered in CVS, it is appropriate to analyze both cytotrophoblasts and mesenchymal tissue. We have previously advocated this because there are cases of true fetal mosaicism (TFM) or clinically significant uniparental disomy where the abnormal cell line is only identified in cytotrophoblasts and not mesenchyme, and vice versa.[9] The identification of an increased risk for very low birthweight in cases with CPM3 further justifies the need. We also agree with Toutain et al. that additional fetal ultrasound surveillance is needed when CPM3 is observed (about 13% of all CPM cases). Currently, it is not possible to provide a risk based on the specific chromosome involved.

We do not believe the association provides any additional justification for the use of cell-free DNA (cfDNA) for RATs as a screening test for FGR. It adds another level of complexity in interpretation. cfDNA RAT screening cannot distinguish between CPM1, CPM3, and TFM. Therefore, to refine the risk for severe low birthweight based on the CPM subgroup would require a CVS in RAT-positive cases. This would be in addition to the amniocentesis that is indicated to help rule out TFM. We have previously pointed out that the use of cfDNA screening for the specific purpose of identifying FGR would be ineffective.[4,5] It also does not meet the requirements of a well-defined disorder with a follow-up definitive and timely diagnostic procedure and is fraught with difficulties because of the alternative explanations for a cfDNA result positive for a RAT.

Finally, we wish to congratulate Toutain et al. for their almost 10-year odyssey to bring attention to the importance of the CPM3 subtype.

## SUPPLEMENTARY INFORMATION

The online version of this article (https://doi.org/10.1038/s41436-019-0665-0) contains supplementary material, which is available to authorized users.

## DISCLOSURE

P.B. is consultant and holds stock options in Natera, Inc. F.R.G. is a full-time employee of TOMA laboratory, Impact Lab Group, without ownership shares. F.R.G. is an expert panel member for Roche and consultant for Menarini Biomarkers. J.F. declares no conflicts of interest.

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