Reply

We are grateful to Drs Monni and Iuculano for their critical scrutiny of the Guidelines on invasive procedures in obstetrics, which was recently published in this Journal on behalf of the ISUOG Clinical Standards Committee (CSC)[1]. This document was written by a group of experts in the field of prenatal diagnosis and then underwent external independent review by a panel of referees from different countries before a final review by the ISUOG Board. The rationale for developing the Guidelines was that, despite a reflex decrease in the number of invasive diagnostic procedures after the introduction of non-invasive prenatal screening (NIPS), it is likely that invasive prenatal diagnosis will not become obsolete any time soon, at least not until new non-invasive platforms address satisfactorily the problem of significant pathogenic chromosomal aberrations other than the common aneuploidies[2].

It is exactly this that is at the core of the Correspondence by Monni and Iuculano, who endorse the necessity and the spirit of these Guidelines but express concerns regarding some of the technical aspects and need for more detailed instructions. To this end, we must clarify that the ISUOG CSC developed these Guidelines on invasive procedures[1], focusing on the methods most commonly used and striving to provide evidence-based recommendations, when possible. In order to address satisfactorily as many as possible of the practitioner's questions, the CSC Task Force actively sought evidence for all aspects of diagnostic prenatal procedures, some of which had never been covered by previously published guidelines, including maternal counseling, asepsis, analgesia, Rhesus-D prophylaxis and post-procedure instructions. On the other hand, similar to the practice followed by all Colleges and Societies developing guidelines, we deliberately chose to avoid binding statements when there was insufficient high-quality evidence to support them – which is, unfortunately, common

in the case of diagnostic procedures. We hope this clarifies the process and reassures the authors regarding the international collaborative approach to the Guidelines.

With respect to the specific concerns expressed by Monni and Iuculano:

- Regarding the exact caliber of needle to be used for amniocentesis, as clarified in the Guidelines[1], non-randomized evidence has shown that the fetal loss rate is similar when a 20-G or 22-G needle is used[3,4]. A plausible explanation for this is that the potential benefit of less trauma using a finer needle is compensated by the longer time needed for fluid retrieval. This is also applicable to chorionic villus sampling (CVS). We cannot therefore recommend that a 22-G needle is used in all cases and do not feel that this is supported by the literature. Moreover, there are no data indicating a preferable technique or needle size, either in singleton or in multiple pregnancies, for genetic pathology testing rather than karyotyping. It is self evident that, in the event of a known history of genetic/Mendelian disease, earlier diagnosis (i.e. CVS rather than amniocentesis) is advisable.
- Regarding timing of transabdominal CVS, the Guidelines advise not performing it before 10 weeks[2]; no upper time limit is specified, as, unlike the transcervical approach, the transabdominal one appears feasible and safe also late in gestation as it does not disrupt the integrity of the cervical barrier.
- The technique of needle insertion is also described, placing emphasis on the possible differences in terms of safety and efficacy of the various approaches (e.g. free-hand *vs* biopsy adaptor) and of different needle calipers. There are technical aspects for which there is no comparative evidence: for example, syringe aspiration *vs* vacuum aspiration or the angle of approach. Some of these aspects are discussed marginally as they fall largely outside the scope of the Guidelines which, by definition, is not a 'How to' article.
- Monni and Iuculano comment on the lack of clear guidance about the optimal technique of fetal sampling according to the gestational age and the indication for testing; in fact, the choice of the most suitable procedure in each case is dependent largely on when it becomes indicated and is further influenced by individual conditions applying both to the patient (e.g. placental location) and the operator (experience with a particular procedure), the particular sonographic findings and even the legal context of each country; all these factors are discussed in the Guidelines. In this regard, although not mentioned by Monni and Iuculano, we take the opportunity to endorse the advice of the International Society for Prenatal Diagnosis, that the confirmatory test after an abnormal NIPS result should preferably be amniocentesis rather than CVS, as placental mosaicism may be found at CVS after a positive NIPS result, and amniotic fluid sampling is ultimately warranted to differentiate a confined placental anomaly from one extending to the fetus[5].
- Regarding the rate of sampling failure at amniocentesis, there are no clear data, as fluid retrieval may be achieved by repeat punctures and this has been acknowledged as a possible risk factor for fetal loss. Moreover, the 2.5–4.8% failure rate of transabdominal CVS that appears in the literature does not take into account experienced referral centers like that of Monni and Iuculano, in which risk of an unsuccessful procedure is expected to be much lower.
- As there is no published evidence on transabdominal CVS *vs* fetal blood sampling in cases with oligohydramnios, we believe that individual conditions and skills should dictate which is the preferred test under these specific circumstances. Of course we agree that it is reasonable to consider the safer and technically easier approach and that placental sampling in such cases seems to pose a smaller risk compared with any procedure entering the amniotic cavity.
- Regarding CVS *vs* amniocentesis in twins, we agree that CVS is preferable in dichorionic twins due to the lower risks of early termination, and this has already been acknowledged in the specific ISUOG Guidelines on twin pregnancy[6]. As for the comment regarding colorant use if amniocentesis is performed by a non-expert, we reaffirm that invasive procedures in twins should be performed preferably by an expert who can also proceed to selective termination, if needed[1]. This also applies to the issue of single uterine puncture and transmembrane sampling *vs* double puncture.
- Monni and Iuculano express their concerns about the lack of recommendation on double sampling if monochorionic twins are discordant for crown–rump length or nuchal translucency thickness; in fact the Guidelines specify that 'sampling of a single sac is warranted when … fetal growth and anatomy are concordant; if this is not the case, double sampling should be considered'[1]. The case of monochorionic twins with twin–twin transfusion syndrome (TTTS) is not discussed in these Guidelines, as the development of TTTS entails the adoption of specific clinical strategies, which are illustrated in the dedicated ISUOG Guidelines on twin pregnancy[6].
- As mentioned in the Guidelines[1], the reason for double sampling in monochorionic twins after *in-vitro* fertilization is the theoretical risk for heterokaryotype, and on this point we endorse the recommendation of the Canadian Prenatal Diagnosis Committee[7], as cited at the end of the corresponding paragraph.
- Monni and Iuculano also raise the issue of written *vs* oral consent. Due to the potential medicolegal implications, the ISUOG Guidelines clarify that 'at the end of this detailed informative process, written consent should be obtained from the woman'[1]. In contrast to the concept 'that the technique used should be the cheapest and the simplest one', as suggested by Monni and Iuculano, our Guidelines, in line with similar national

and international guidelines, emphasize the importance of tailoring the procedure to the context, including local conditions and resources.

- We welcome the authors' input on optimal training strategy, which highlights the need that training should start in a way that it is comfortable and profitable to the trainees and safe to the pregnant women.

Once again, we thank the authors for their comments. They work within a team with vast clinical and research experience on the subject and have raised many important points. We hope our response addresses their questions and highlights the specific points of practice for professionals who are engaged in prenatal diagnosis.

T. Ghi*†, A. Sotiriadis‡ and N. Raine-Fenning§
†Department of Obstetrics and Gynecology, University of Parma, via Gramsci n 14 Parma, Parma 43126, Italy;
‡EMVRYO PCC, Thessaloniki, Greece;
§Division of Child Health, Obstetrics and Gynaecology, School of Medicine, University of Nottingham, Nottingham, UK
**Correspondence.*
(e-mail: tullioghi@yahoo.com)
DOI: 10.1002/uog.17376

References

1. Ghi T, Sotiriadis A, Calda P, Da Silva Costa F, Raine-Fenning N, Alfirevic Z, McGillivray G; International Society of Ultrasound in Obstetrics and Gynecology (ISUOG). ISUOG Practice Guidelines: invasive procedures for prenatal diagnosis. *Ultrasound Obstet Gynecol* 2016; **48**: 256–268.
2. Evans MI, Wapner RJ, Berkowitz RL. Noninvasive prenatal screening or advanced diagnostic testing: caveat emptor. *Am J Obstet Gynecol* 2016; **215**: 298–305.
3. Athanasiadis AP, Pantazis K, Goulis DG, Chatzigeorgiou K, Vaitsi V, Assimakopoulos E, Tzevelekis F, Tsalikis T, Bontis JN. Comparison between 20G and 22G needle for second trimester amniocentesis in terms of technical aspects and short-term complications. *Prenat Diagn* 2009; **29**: 761–765.
4. Uludag S, Aydin Y, Ibrahimova F, Madazli R, Sen C. Comparison of complications in second trimester amniocentesis performed with 20G, 21G and 22G needles. *J Perinat Med* 2010; **38**: 597–600
5. Benn P, Borrell A, Chiu RW, Cuckle H, Dugoff L, Faas B, Gross S, Huang T, Johnson J, Maymon R, Norton M, Odibo A, Schielen P, Spencer K, Wright D, Yaron Y. Position statement from the Chromosome Abnormality Screening Committee on behalf of the Board of the International Society for Prenatal Diagnosis. *Prenat Diagn* 2015; **35**: 725–734.
6. Khalil A, Rodgers M, Baschat A, Bhide A, Gratacos E, Hecher K, Kilby MD, Lewi L, Nicolaides KH, Oepkes D, Raine-Fenning N, Reed K, Salomon LJ, Sotiriadis A, Thilaganathan B, Ville Y. ISUOG Practice Guidelines: role of ultrasound in twin pregnancy. *Ultrasound Obstet Gynecol* 2016; **47**: 247–263.
7. Audibert F, Gagnon A; Genetics Committee of the Society of Obstetricians and Gynaecologists of Canada; Prenatal Diagnosis Committee of the Canadian College of Medical Geneticists. Prenatal screening for and diagnosis of aneuploidy in twin pregnancies. *J Obstet Gynaecol Can* 2011; **33**: 754–767.