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Role of Chorionic Villous Sampling in prenatal diagnosis Our experience.

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Abstract : Introduction-Chorionic villous sampling(CVS) is an invasive method of prenatal diagnosis to determine chromosomal or genetic disorders in the fetus.It is performed between 10-13 weeks, either transvaginally or tranabdominally .The primary advantage of CVS is that results are available earlier in pregnancy. The complications of CVS are fetal loss (0.7) within 14 days, amniotic fluid leakage and infections (0.5).CVS performed earlier than 9 weeks is associated with limb reduction. The aim of this study is to analyse the feasibilty of the procedure and to evaluate the indications, results and complications of trans abdominal chorionic villus sampling in prenatal diagnosis. Material Method This is a descriptive analytical study conducted in the Perinatal medicine clinic in the Department of Obstetrics and Gynaecology Unit 4 ,at CMC Vellore over a period of one year, from August 2014- August 2015. Data of women undergoing transabdominal CVS for various indications were obtained from USG room record.. Indication of the procedure as well as results, complications, technical failure and maternal contamination of the sample were analysed. Results Totally 53 patients underwent CVS during the study period . The mean maternal age was 29.09yrs and mean gestational age was 12 weeks and 3 days. The various indications as per analysis were 24(45.2) to rule out chromosomal abnormality,20(37.7)to rule out genetic disease, and 9(16.9) to rule out haematological disorder. Results of the CVS were analysed in the chromosomal abnormality group, normal results were found in 15(62.5) patients, abnormal results were found in 8(33.3) patients. In the genetic disease group , the fetus was not affected in 14(70),fetuses were affected in 6(30).In the haematological disorder group, fetus not affected in 4(44.4) patients ,affected foetuses were found in 3(33.3) patients, and fetus with carrier state was found in 1 (11.1) patient. Maternal contamination was found in 3(5.6) samples. CVS was unsuccessful in 2 (3.6)patients due to technical difficulty. Conclusion CVS is a safe and reliable prenatal diagnostic technique. It should be one of the options available to pregnant women who require prenatal diagnosis.
Keyword : Chorionic villous sampling , prenatal diagnosis , indications, outcome.

Introduction:
Invasive prenatal diagnosis consists of a set of instrumental and laboratory techniques that permit diagnosing genetic, chromosomal, metabolic and infective pathologies (1).The most widely used invasive prenatal procedures currently used are: chorionic villous sampling(CVS), amniocentesis and fetal blood sampling by cordocentesis(2).All these techniques are performed under continuous ultrasound guidance. Recently, CVS is becoming more widely used compared to the rest of the techniques. Its advantage is that it can be performed at an early stage of pregnancy mitigating the physical and psychological problems correlated to a late diagnosis(3,4).Various modes of sampling exist: transabdominal, transcervical and transvaginal. The transcervical and transvaginal modes can be performed until 13th week of gestation whereas the transabdominal method has the advantage that it can be performed also in the 2nd and 3 rd trimester(5). The first CVSs were employed for diagnosing Mendelian pathologies (6), followed closely by chromosomopathies(7).In recent years, due to the introduction of first-trimester screening tests for aneuploidies, CVS for karyotyping has become largely diffused (8). Main indications for chorionic villous sampling:1.Advanced maternal age2. Positive ultrasound and biochemical first -trimester aneuploidy screening 3.Parents with chromosomal anomalies 4.Previous child with chromosomal anomaly 5. Previous child with plurimalformations 6.High risk of single-gene disorders 7.Oligo/anhydraminos 8.Fetal anomalies detected by ultrasound 9.X-linked disease 10.Infectious diseases(TORCH) (10-12). Contraindications of the procedure are maternal alloimunisation, HIV ,hepatitis B and C,bleeding or spotting per vaginum and acute ante/retroversion of the uterus. The aim of this study is to analyze the feasibilty of the procedure and to evaluate the indications, results and complications of trans abdominal chorionic villus sampling in prenatal diagnosis.

Material & Method : This is a descriptive analytical study conducted in the Perinatal medicine clinic in the Department of Obstetrics and Gynaecology Unit 4 ,at CMC Vellore over a period of one year, from August 2014- August 2015. Data of women undergoing transabdominal chorionic villous sampling for

various indications were obtained from the ultrasound(USG) room data base .The samples were analysed in cytogenetic,haematological and medical genetics labs. Maternal characteristics such as age, gestational age, parity,domicile were looked into. Indication of the procedure as well as results, complications, technical failure and maternal contamination of the sample were also analysed.

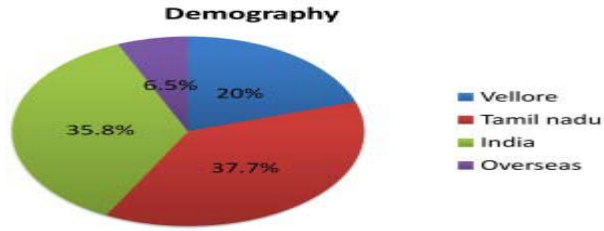
Results:

In this study 53 patients were included during the study period,CVS were done for various indications, 12.7% for chromosomal abnormalities,25.4% was done for genetic disease and 10.9% was done for haematological disorders .Of those who had CVS for chromosomal abnormality,34.5% was for those who tested screen positive for trisomy and 12.7% had previous child with chromosomal abnormality,11.2 % for family history of genetic disease. Among the patients who had CVS for haematological disease 10.9% were done for previous affected child and 5.4% for parents with carrier status.

Analysis of indications for the CVS in the study:

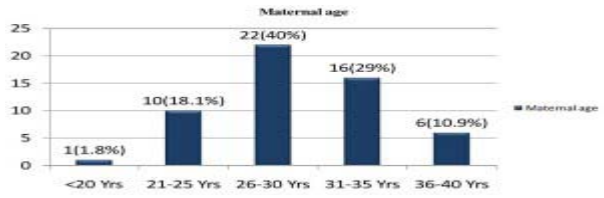
Demographic distribution of the patients:

Among the 53 patients, 37.7% were from Tamil Nadu excluding vellore,35.8% patients from rest of India,18.8% patients from vellore, and 7.5% patients from overseas.



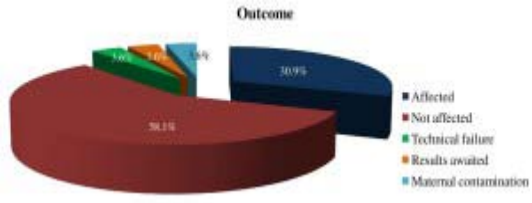
Maternal age distribution:

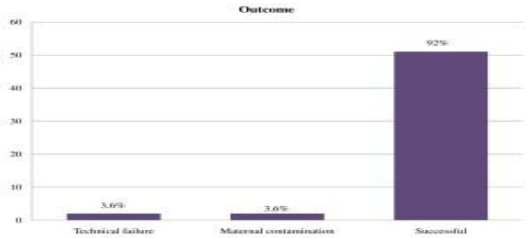
Among 53 patients , 40% were between 26-30yrs, 29% were 31-35 yrs ,18.1% were 21-25yrs, 10.9% were between 36-40 yrs ,1.8% were less than 20yrs.



Outcome:

Prenatal diagnosis - Indication	Affected	Not Affected	Technical failure	Maternal contamination	Results awaited	Total-55(%)
Aneuploidy	8	15	2	1	0	26(47.2%)
Genetic syndromes	5	13	0	-	2	20(36.6%)
Haematological disorders	4	4	0	1	0	9(16.3%)





Among 53 patients, 92% of the procedures were successful ,58.1% had normal outcome, 30.9% were affected 3.6% of patients had technical failure and 3.6% of sample had maternal contamination.

Discussion:

CVS is the gold standard for first trimester prenatal diagnosis. In the study done by Oa Olayede et al the various indications of CVS in the first trimester were analysed , and the most common indications were for haematological disorder and aneuploidy and most procedure were done by transabdominal route and 98% of the procedure was successful(39) The results were similar to our study

Counselling and consent before the procedure :

Non –directive genetic counselling must be carried out by doctors/ experts in obstetrics and /or genetics before the procedure (13). Such counselling must provide all necessary information so that the couple can make their choice concientiously. The correct information on the gestational age, sampling procedure and its mode should be provided as well as the possibility of sampling procedure failure and the necessity of repeating the procedure in case of inadequacy of the sample quantity. The risk of fetal loss and the possibility for performing alternative invasive diagnosis technique such as amniocentesis or cordocentesis must also be discussed (14).Whether the women decides to undergo the procedure or not ,written or oral consent must be obtained with all the provided information specified.

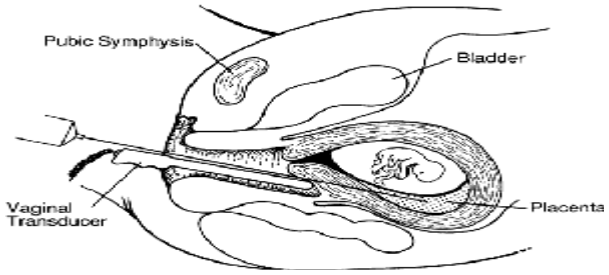
Techniques:

Various method of performing CVS exist. The access can be transcervical, transvaginal or transabdominal. Transabdominal route is currently the most largely employed. All procedures are performed under continuous ultrasound guidance. An integral part of any specific mode is the preliminary ultrasound study of the pregnancy for establishing the viability, number of foetuses, biometry, chorionic position and chorionicity in case of twin or multiple pregnancies. The best chorionic zone to insert the needle is the one that allows the shortest route to reach the sampling spot and also avoids possible contracted areas, fibroids, areas of deciduous chorionic detachment and haematomas(15). The possibility and the utility of using ultrasound 3D or 4D for both transabdominal and transcervical sampling is being studied for several years(16).The theoretic utility is undisputed as the visualisation of the three levels of scanning helps the identification of the cannula tip or of the needle mostly when it is difficult to follow .

Transcervical sampling :

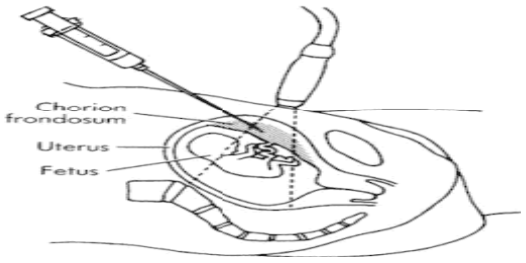
Transcervical sampling can be performed by aspiration via cannula or catheter of good flexibility and manageability or using biopsy rigid or curved forceps of about 2-mm diameter(17). During the procedure the assistant must continually offer information of the relation between the chorion, the cervical canal and the sampling zone by transabdominal ultrasound guidance. The aspiration of the villous sampling requires of a 16-gauge cannula on the external orifice of the uterus. Once introduced, the cannula points must be visualised on the ultrasound by the assistant so as to permit the operator to reach the inferior placenta margin and the chorion frondosum. On reaching the sampling spot, a syringe containing culture liquid, about 5 ml, and heparin must be connected at the distal end and negative pressure should be applied, moving the cannula several times on its longitudinal axis. When the villous sampling is obtained, the cannula must be removed with the syringe (18). The sampling by biopsy forceps requires the insertion of the forceps inside the external orifice of the uterus and the concurrent visualisation of the point in the monitor; the introduction must be far from the deciduous chorionic junction(19).Once it is reached ,the operator samples the villi and the biopsy forceps is removed cautiously. With this mode of sampling, it is important to visualise the point of the forceps immediately after it is introduced at the level of external orifice of the uterus so as to avoid creating a false route. The transcervical sampling has some absolute contraindication such as current vaginal infections, vaginism and acute cervical stenosis whereas the presence of fibroid in the lower part of the uterus can sometimes hinder the introduction of the needle(18).

Transvaginal Sampling Technique



The transvaginal technique requires the use of a vaginal probe with the special guiding needle. The women must be prepared as in the case of transcervical mode of sampling and the probe must have a sterile wrapping. Once having introduced the probe with the special guiding needle and chosen the sampling spot, the needle must be introduced to cross the vagina, the parametrium and the uterine wall until it reaches the chorion frondosum. It should be moved back and forth. The technique is not widely employed as it does not present special advantage compared to the other two techniques. Its use has been suggested in cases of failure of the other two sampling modes and nowadays it is almost completely abandoned(20).

Tranabdominal Sampling Technique



The abdominal sampling can be performed with a double coaxial needle or a single one. The recommended stage is usually 11th-12th weeks. The double needle is fixed at one end of the transducer. Once the spot for villous sampling is chosen, the 18-guage needle is introduced until it reaches the myometrium, till the chorion. Meanwhile a second needle,20 or 22 guage ,is introduced inside the first one so as to reach the chorion. The advantage of this is that the direction of the needle is predetermined; hence it reduces possible errors during reaching the chosen spot and at the same time prevents any digression of the direction during the sampling process. This may, however, prove to be a disadvantage(21). The single needle mode does not necessitate the use of a support but is performed free- hand. This technique requires employing a spinal 20-guage needle till the 13th week of pregnancy; For advanced gestation, it is preferable to use a 19 or 18 guage needle(5).The direction of the needle introduction has to be tangential to the ultrasound probe in its central part lengthwise in order to meet the ultrasound waves right after its insertion in the maternal abdomen. After being introduced ,the needle is connected to a 2-ml syringe. The displayed needle tip is moved up and down 5-10 times to favour detachment and suction of villi while applying a negative pressure, by manual aspiration through the syringe(22).This technique, contrary to the previous one, as already pointed out, allows the possibility of changing the direction of the needle once introduced in the abdomen but requires major experience of the operator as the tangential insertion of the needle enables the visualisation of the needle point only(23). With the other free-hand technique employing moving “up and down” aspiration ,we can use a syringe of 2.5ml or a pistol connected to a 20-30ml syringe .With this technique, the needle is used in oblique position to the ultrasound probe. This mode has the advantage of entirely visualising the needle during the introduction in the chorion but necessitates a longer route and is thus more painful for the woman(24). The transabdominal route can be performed by a single operator who holds, simultaneously, the probe and the needle so as to optimise the monitor visualisation and requires the help of an assistant just for connecting the needle to the syringe once the sampling spot is reached. Another technique is the “three- hand” sampling ,that is ,with an assistant holding the ultrasound probe and the operator connecting the needle to the syringe. A technique which has been recently proposed for single-needle sampling is transabdominal CVS with vacutainer(25).This mode requires a single operator and the use of 4 ml vacutainer which, once connected to the needle ,exerts constant negative pressure and facilitates the sampling process. The contra-indications of both transcervical and transabdominal techniques are the presence of vaginal bleeding and big-sized haematomas. Rh-negative patients with a negative coombs test must usually receive the necessary prophylaxis by administering immunoglobulins anti-D as the CVS may provoke Rh isoimmunisation(26). In HIV ,Hepatitis B and C there were significant rates of transmission where no treatment was in place (25%) and where mono or double therapy was used (6.1%).Whenever possible, procedures should be delayed until treatment has optimised the maternal viral load (37).Evidence does not suggest administration of antibiotics or tocolytic as a routine. The antibiotic prophylaxis is indicated to women with cardiac valve anomalies. In case of a completely posterior placenta and a retroverted uterus,it is possible to perform a simple

manipulation of the cervix by the assistant pressing the anterior fornix. This manipulation helps to improve the position of the trophoblast in cases of difficult sampling procedure. Neither local nor general anaesthetic is necessary. Randomised trials confirm that anaesthesia does not automatically imply the reduction of pain perception (27).Therefore ,it is normal that during the sampling procedure the women perceives pain mainly at the moment when the needle penetrates the uterus; the pain usually subsides shortly after the end of the procedure. A study conducted in 2009 (28) reveals that the transabdominal sampling mode is more painful for women with major body mass index and the transcervical one more painful for the nullipara ones. Sometimes repeating the sampling procedure can prove necessary due to : insufficient material, wrong introduction of the instrument and the formation of a clot inside the needle which obstructs the aspiration of chorionic villi. It is not recommended to perform the sampling procedure more than two times per session. The third sampling ,in fact, increases the fetal loss risk considerably and hardly proves successful after having tried twice(29).It is useful to postpone the sampling procedure even by a whole week in such cases.

Complications:

Major and minor complication after CVS

	Spotting	Bleeding	Infection	Amniotic fluid loss	Fetal loss
TV-CVS	1/3	Rare	1/1000	0.5%	0.5-1%
TA-CVS	<1/3	Rare	1/1000	0.5%	0.5-1%

Among the minor complications, the most frequent is bleeding which is present in about one-third of the patients who have been tested by the transcervical sampling procedure and less than one-third of the patients who have been tested by the transabdominal one. Abundant bleeding is a less common event but more frequent with transcervical sampling procedures. Infection and amniotic liquid loss are present in 0.5% of patients. The chorioamnionitis risk is low, less than 1:1000(30). Regarding the major complications, fetal loss after CVS is sometimes difficult to quantify. The spontaneous abortion incidence at 11-12 weeks of gestation is in fact about 1% in the general population. It must be considered that women who undergo the CVS procedure are usually women at higher pregnancy risk and therefore at higher risk for spontaneous abortion. Considering this, the postprocedure abortion risk is estimated to coincide with the one presented by amniocentesis in the second trimester :about 0.5% (31,32).A Cochrane library publication of 2003 reports abortion risk after TC-CVS to be higher than in cases of TA-CVS(33).Factors which expose to higher risk for fetal loss are: more than two needle insertions per sampling procedure, the presence of considerable blood loss during the sampling procedure, advanced maternal age (older than 35 years), gestational period inferior to 10 weeks (34),presence of haematomas which increases the risk for spontaneous abortion. Maternal obesity increases the abortion risk after amniocentesis but not CVS(35). Other major complication after a CVS which are reported are limb and oro-mandibular (mandibular hypoplasia)defects(36).The increase incidence of limb defects after CVS is controversial. The World health Organisation reports incidence equal to 6/1000 cases and such risk is not significantly different than the risk in the general population (37,38).However,various studies show a slight incidence of limb defects when the CVS is performed before the 9th gestational week, other studies mention the 7th week(30).

CVS in Twin pregnancy:

A high level of expertise in ultrasound scanning is essential for operators undertaking amniocentesis or CVS in multiple, dichorionic pregnancies, because uterine contents have to be 'mapped' with great care. This is essential to ensure that separate samples are taken for each fetus and clearly labelled as such. The miscarriage rate is likely to be higher than in singleton pregnancies. A recent singlecentre study of 311 twin, mid-trimester amniocenteses estimated the attributable pregnancy loss rate at 1/56 (1.8%)(38). The role of CVS in dichorionic placentas remains controversial because of a relatively high risk of crosscontamination of chorionic tissue, which may lead to false positive or false negative results. This risk may be minimised if two separate needles are used. Such procedures should be performed only after detailed counselling.

Conclusion:

CVS is a relatively safe and reliable method of prenatal genetic diagnosis. The acceptance and utilization of CVS is beginning to increase especially among pregnant women. Transabdominal and transcervical chorionic-villus sampling appear to be equally safe procedures for first-trimester diagnosis of fetal abnormalities. Hence we can offer this procedure to our patients safely.

References :

1.Milunsky A, Milunsky JM.*Genetic Disorders and the fetus:Diagnosis ,prevention and Treatment* (6th edn.).UK:wiley-Blackwell;2010.

2.Dalton ME,De cherney AM.Prenatal diagnosis.*N Engl J Med*.1993;328:114-20

3.Cederholm M,Axelsson O,Sjoden PO.Women's Knowledge, concerns and psychological reactions before undergoing an invasive procedure for prenatal karyotyping.*Ultrasound ObsetGynecol* .1999;14:267-72

4.Cao A,Cossu P,Monni G,Rosatelli C.Chorionic villous sampling and acceptance rate of prenatal diagnosis.*Prenat Diagn* 1987;7:531-3

5.Monni G.Ibba RM,Olla G ,Rosatelli C Cao A.Prenatal diagnosis of -thalassemia by second trimester chorionic villus sampling. *Prenat Diagn.*1988;8:447-51.

6.Ward RH,Model B,Petrou M,Karagozlu F,Douratsos E.Method of sampling chorionic villi in first trimester of pregnancy under guidance of real time ultrasound.*BMJ*.1983;286:1542-4

7.Simoni G,Brambati B,Danesino C,*et al*.Efficient direct chromosome analysis and enzyme determinations from chorionic villi samples in the first trimester of pregnancy.*Hum Genet* .1983;63:349-57.

8.Pandya PP, Brizot ML, Kuhun P, Snijders RJM, Nicolaides KH.first –trimester fetal nuchal translucency thickness and risk for trisomies. *Obstet Gynecol.* 1994;84:420

9.Oztekin O, Oztekin D, Tinar S,Adibelli Z.Ultrasonographic diagnosis of fetal structural abnormalities in prenatal screening at 11-14 weeks.*Diagn Interv Radiol*.2009;15:221-5

10.Bosma TJ,Corbett KM,Eckstein MB,*et al.* Use of PCR for prenatal and postnatal diagnosis of congenital rubella.*J Clin Microbiol*.1995;33:2881-7

11.Kustermann A,Zoppini C,Tassis B, *et al*. Prenatal diagnosis of congenital Varicella infection. *Prenat Diagn*.1996;16:71-4

12.Dong ZW,Zhou SY,LiY *et al.* Detection of a human parvovirus intrauterine infection with polymerase chain reaction.*J Reprod Med*.2000;45:410.

13.Milunsky A,Milunsky JM.Genetic counselling: preconception, prenatal and perinatal.*Genetic disorder and the Fetus:Diagnosis,Prevention and Treatment*(6th edn.).UK:wiley-Blackwell;2010:1-62.

14.Dodd JM,Crowther CA.Misoprostol for induction of labor to terminate pregnancy in the second or third trimester for women with afetal anomaly or after intrauterine fetal death.*Cochrane Database Sys Rev*.2010;14

15.Monni G,Zoppi MA,Axiana C.Chorionic villus sampling.In:*Clinical Maternal-Fetal Medicine* (Chapter

62).Winn Hn,Chervenak FA,Romero R(eds).USA:Informa Healthcare;2011.
16.Adeniji B,Williams J,III,Solt I,Morales C,Alanakian A,Rotmensch S.Clinical trial of multiplanar realtime 4- versus 2-dimensional sonographic guidance for transcervical chorionic villus sampling.*J Ultrasound Med*.2011;30:309-12
17.Alfirevic Z,von Dadelszen P.Instrument for chorionic villus sampling for prenatal diagnosis. *Cochrane Database Syst Rev* 2003;(1)
18.Brambati b.chorionic villus sampling.*Curr Opin Obset Gynecol*.1995;7:109-16
19.Monni G,Ibba RM,Olla G,Rosatelli C ,Cao A.Chorionic villus sampling by rigid forceps:experience with 300 cases at risk of thalassemia major.*Am J Obset Gynecol.*1987;156:912-4
20.Sidransky E,Black SH,SoenKsen DM,Jones SL,Dorfmann AD,Schulman JD. Trans-vaginal chorionic villus sampling.*Prenat Diagn*.1990;10:583-6.
21.Smidt-Jensen S,Hahnemann N.Transabdominal fine needle biopsy from chorionic villi in the first trimester. *Prenat Diagn*.1984;4:163-9
22.Monni G,Ibba RM,Lai R,*et al*.Early trasabdominal chorionic villus sampling for couples at high genetic risk.*Am J Obset Gynecol*.1993;168:170-3
23. Monni G,Ibba RM,Zoppi MA.Prenatal genetic diagnosis through chorionic villus sampling.*In: genetic Disorder and the Fetus:Diagnosis,Prevention and Treatment* (6th edn).Milunsky JM (eds).UK:Wiley-Blackwel;2020:160-93
24.Brambati B.Oldrini A Lanzani A.transabdominal chorionic villus sampling: a freehand ultrasound guided technique.*AM J Obset Gynecol*.1987;157:134-7.
25.Battagliarin G ,Lanna m,Coviello D,Tassi S.Quarenghi A,Nicolini U.A randomised study to assess two different technique of aspiration while performing transabdominal chorionic villus sampling. *Ultrasound obstetric Gynecol*.2009;33:169-72.
26.American college of Obstetricians and Gynecologist Committee on Practice Bulletins- Obstetrics.ACOG Practice Bullettin.Clinical management guidelines for Obstetriciangynecologist. Prenatal diagnosis of fetal chromosomal abnormalities.*Obset Gynecol.*2001;97:1
27.Vandenbossche F,Horovitz J,Guyon F,Verret C,Saura R.Pain experience during chorionic villus sampling and amniocentesis: a preliminary study.*Eur J Obset Gynecol reprod Biol*.2008;136:189-93.
28.Wax JR,Davies NP,Watson WJ,*et al* Pain associated with chorionic villus sampling:transabdominal vs transcervical approach.*Am J Obset Gynecol*.2009;201-400e1-3.
29.Tabor A,Alfirevic Z.Update on procedure-related risks for prenatal diagnosis techniques.*Fetal Diagn Ther*.2010;27:1-7.
30.Brambati B,Tului L.Prenatal genetic diagnosis through chorionic villous sampling.In:*Genetic Disorder and the Fetus:Diagnosis,prevention ,and Treatment*(5th edn.).Milunsky A (ed.).Baltimore,MD:Johns Hopkins University Press;2004;179-213.
31.Multicentre randomized clinical trial of chorionic villus sampling and amniocentesis.First report.Canadian Collobrative CVS-Amniocentesis Clinical Trial Group.Lancet.1998;351:242-7.
32.Mujeziinovic F,Alfirevic Z.Procedure-related complication of amniocentesis and chorionic villous sampling: a systematic review.*Obstet Gynecol*.2007;110-687-94.
33.Alfirevic Z,Sundberg K,Brigham S.Aminicentesis and chorionic villous sampling for prenatal diagnosis.*Cochrane Database Syst Rev*.2003;3:CD003252
34.Monni G,Ibba RM,Lai R,*et al* .Transabdominal chorionic villus sampling:fetal loss rate in relation to maternal and gestational age.*Prenal Diagn*.1992;12:815-20
35.Harper LM,Cahill AG,Smith K,Macones GA,Odibo AO.Effect of maternal obesity on the risk of fetal loss after amniocentesis and chorionic villus sampling .*Obstet Gynecol*.2012;119:745-51.
36.Firth HV,Boyd PA,Chamberlain P et al, Severe limb abnormalities after chorionic villus sampling at 56-66-days'gestation.*Lancet*.1991;337:762-3.
37. Mandelbrot L, Jasseron C, Ekoukou D, Batallan A, Bongain A, Pannier E, et al. Amniocentesis and mother-to-child transmission of human immunodeficiency virus in the Agence Nationale de Recherches sur le SIDA et les Hepatites Virales French Perinatal Cohort. Am J Obstet Gynecol 2009;200:e1–9
38.Cahill AG, Macones GA, Stamilio DM, Dicke JM, Crane JP, Odibo AO. Pregnancy loss rate after mid-trimester amniocentesis in twin pregnancies. Am J Obstet Gynecol 2009;200:257
39.Clinical and laboratory experience of chorionic villous sampling in Nigeria-2014,Nigerian journal of clinical practice.