Reference interval for fetal biometry in Italian population

Maurizio Giorlandino[1], Francesco Padula[1], Pietro Cignini[1], Marialuisa Mastrandrea[1], Roberto Vigna[1], Giorgia Buscicchio[2], Claudio Giorlandino[1]

[1] "Artemisia" Fetal-Maternal Medical Centre, Department of Prenatal Diagnosis, Rome, Italy
[2] Department of Obstetrics, Marche Polytechnic University, Ancona, Italy

Corresponding Author: Claudio Giorlandino, MD
Fetal-Maternal Medical Centre ARTEMISIA
Department of Prenatal Diagnosis
Viale Liegi, 45 - 00198 Rome, Italy
Phone: +39 068505250/+39 068505
Fax: +39 068505280
E-mail: claudio.giorlandino@artemisia.it

Abstract

Objectives: **To validate new references charts and equations for fetal biometry in an Italian unselected population.**
Methods: **A cross-sectional study involving 4896 women with singleton viable pregnancies, at Artemisia Fetal Maternal Medical Centre between May 2009 and December 2009. Each woman was scanned only once, between 14^{+0} and 40^{+0} weeks of gestation. The fetal standard biometric measurements were recorded. For each parameter, regression models were fitted to estimate the percentile at each gestational age. In order to be compared to other reference equations, the fetal biometric measurements at each gestational age were expressed as Z-scores.**
Results: **New fetal charts and references equations for Italian population were developed according to the recommend multistep statistical procedure.**
Conclusion: **To our knowledge this is the first Italian study with the largest sample size ever reported in the literature. In addiction, our newer charts of reference centiles for fetal biometric measurements are useful in the obstetrical clinical practice for the Italian population.**

Introduction

An appropriate evaluation of fetal biometry represents the cornerstone in the fetal growth assessment giving that an abnormal growth may be associated with an adverse perinatal outcome and may require a specific obstetrical care[1].

Many variables affect fetal growth[2, 3, 4] including physiological and pathological changes, such as maternal height and weight, drug or tobacco exposure, fetal sex, ethnicity[5], genetic syndromes, congenital anomalies and placental failure. Therefore, each particular population or ethnic group should have their own reference charts for the different fetal anthropometrical variables in order to provide the most accurate fetal assessment. Furthermore, fetal nomograms need to be revised regularly as fetus is growing up in the last decades[6, 7] and need to be constructed in accordance with the recommended method of analysis[8, 9]. In fact, the choice of a standard approach is crucial, because inaccurate centiles obtained from an inferior method may mislead the obstetrician as to the true state of health of development of the fetus and increase the chance of suboptimal clinical care. The World Health Organization recommends the expression of measurements as Z-scores in order to allow relevant statistical analysis[10, 11].
The aim of our study is to develop new reference charts and equations for fetal standard biometry between 14 and 40 weeks of gestation in a cohort of healthy Italian women with low-risk singleton viable pregnancies.

Methods

The study included a population of 4896 Italian women undergoing ultrasound examination between the 14^{th} and 40^{th} weeks of gestation at Artemisia Fetal Maternal Medical Centre, between May 2009 and December 2009. Inclusion criteria were singleton pregnancies in Italian women with low-risk pregnancy. Exclusion criteria were multiple pregnancy, abnormal karyotype, congenital malformations, gestational or maternal diseases (such as hypertension and diabetes mellitus), no first-trimester dating based on crown-rump length (CRL). Cases with low birth weight, preterm delivery or other prenatal complications were not excluded. Gestational age was based on the last menstrual period and in all cases adjusted according to the CRL measured in the first trimester ultrasound. Every fetus was measured only once for the study. All measurements were performed by 5 different obstetric sonographers (MG, PC, FP, MLM, RV) using the following equipments: GE Voluson 730, GE E8 and GE Voluson 730 Pro (General Electric Healthcare, USA).
Measurements of the biparietal diameter (BPD) and head circumference (HC) were obtained from a transverse axial plane of the fetal head showing a central midline echo broken in the anterior third by the cavum septum pellucidi and demonstrating the anterior and posterior horns of the lateral ventricle. The BPD was measured from the outer margin of the proximal skull to the inner margin of the distal skull. The HC was measured fitting a computer-generated ellipse to include the

outer edges of the calvarial margins of the fetal skull. The abdominal circumference (AC) was measured fitting a computer-generated ellipse through a transverse section of the fetal abdomen at the level of the stomach and bifurcation of the main portal vein into its right and left branches. The femur length (FL) was measured in a longitudinal scan where the whole femural diaphysis was seen almost parallel to the transducer and measured from the greater trochanter to the lateral condyle. In the third trimester, particular care was taken not to include the ephiphysis.

Statistical analysis was performed using SPSS 16.0 and STATISTICA 7.0. The data were analyzed as recommended by Altman and Royston [8, 9].

The normality of measurements at each week of gestation was assessed using the Kolmogorov–Smirnov test. Given the large sample size, statistically significant non-normality was accepted unless the normal plot showed clear deviation from a straight line. In order to obtain normal ranges for fetal measurements, a multistep procedure based on regression model has been used, according to the recommended methodology.

A centile curve is calculated using the following formula:

Centile = M + *K* x *SD*

where K is the corresponding centile of the standard Gaussian distribution, M is the mean and SD is the standard deviation of the mean of the fetal measurements for each gestational age.

The mean is estimated by the fitted values from an appropriate polynomial regression curve of the measurement of interest on gestational age. A cubic polynomial model was used ($y = a + b \times GA + c \times GA^2 + d \times GA^3$) unless the R^2 statistic and/or the fitted curve was not satisfactory.

The standard deviation is obtained through the following steps: the residuals were calculated as the absolute value of the differences between the fitted curve and the original data for each patient and then multiplied by a corrective constant equal to (=1.253). These were finally regressed on gestational ages by using a simple linear equation (y = a + b x GA). The fitted values give the age-specific SD estimates.

Finally, these predictive mean and SD equations allow calculating any required centile, replacing the value in the centile formula. The constant K may be ±0.674 for the determination of the 25th and 75th centiles, ±1.282 for the 10th and 90th centiles, ±1.645 for the 5th and 95th centiles, ±1.96 for the 2.5th and 97.5th centiles, ±2.33 for 1st and 99th centiles, ±2.58 for 0.5th and 99.5th centiles.

In order to compare our new reference equations with previously published ones, we calculated Z-scores of the median, 5th and 95th percentiles, according the following formula:

$Z\text{-score} = (X_{GA}-M_{GA})/SD_{GA}$

where X_{GA} is the measured value at a known gestational age (GA), M_{GA} is the mean value obtained with the reference equation used at this GA, and SD_{GA} is the SD associated with the mean value at this GA obtained with the reference equation. Results were presented graphically across GA.

Results

Full biometric measurements (BPD, HC, AC, FL) were obtained for 4896 fetuses. The best-fitted regression model describing the relationship between biometric parameters and GA was the cubic model. Data analysis showed that neither the use of fractional polynomials nor the logarithmic transformation improved the fitting of the curves. The corresponding regression equations for the mean are as follows:

$BPD_{mean}=12.707 - (0.771 \times GA) + (0.186 \times GA^2) - (0.00291 \times GA^3)$ (R^2=96.6)

$HC_{mean}=18.841 + (0.506 \times GA) + (0.576 \times GA^2) - (0.00973 \times GA^3)$ (R^2=97.4)

$AC_{mean}= -40.349 + (7.209 \times GA) + (0.214 \times GA^2) - (0.00383 \times GA^3)$ (R^2=95.2)

$FL_{mean}=-32.045 + (3.597 \times GA) - (0.00917 \times GA^2) - (0.000353 \times GA^3)$ (R^2=97.2)

SDs across GA were fitted using a simple linear model. Fits for SDs were as follows:

$BPD_{SD}= 1.174 + 0.069 \times GA$

$HC_{SD}= 1.795 + 0.295 \times GA$

$AC_{SD}= -1.447 + 0.542 \times GA$

$FL_{SD}= 0.676 + 0.061 \times GA$

Tables 1-4 show the centile charts with 0.5th, 1th, 2.5th, 5th, 10th, 50th, 90th, 95th, 97.5th, 99th, 99.th centiles fitted for all biometric parameters. Figure 1 illustrates the goodness of fit of our model by showing the raw data for the main measurements in obstetrical practice with the fitted 5th, 50th and 95th centiles. We graphically compared our Z-scores to the ones derived by the 5th, 50th and 95th percentiles calculated for the main biometric parameters of two previously published sets of reference equations[12, 13] (Figure 2).

Discussion

The need for reference curves for a specific population arises from the observation that fetal growth depends on many factors, including ethnicity. In fact in literature a variety of chart references are described, varying from different populations. On the other hand, there has been an increasing value of fetal biometric parameters, thus the needing of updated chart references that reflect the trend of fetal growth. The first statistical models developed for Italian fetuses date back to 1987, including biparietal diameter, head circumference and abdomen circumference from 1,426 healthy fetuses[14]. More complete Italian fetal size charts will be published almost 20 years later, including head, abdomen and long bones measurements, although the number of fetuses was only 626 and the Authors did not specify the formula for calculating the standard deviation, letting the comparison with their curves be impossible[15].

The construction of reference interval for fetal variables is crucial as inaccurate centiles produced by inferior methods may mislead an abnormal growth that may be associated with an adverse perinatal outcome and require a specific obstetrical care. We followed the method described by Altman and Chitty and Royston and subsequently confirmed by Royston, in which they claim the need of an approach based on statistical techniques and the term “normal range” should be substituted by “reference interval” since the former implies that any observation outside the range indicates an “abnormality”, which is not necessarily the case.

Table 1 Fitted centiles of biparietal diameter (BPD) (mm)

GA	0,5°	1°	2,5°	5°	10°	25°	50°	75°	90°	95°	97,5°	99°	99,5°
14	24,9	25,4	26,2	26,9	27,6	28,9	30,4	31,8	33,1	33,9	34,6	35,4	35,9
15	27,5	28,0	28,8	29,5	30,3	31,7	33,2	34,7	36,0	36,8	37,5	38,3	38,9
16	30,2	30,8	31,6	32,3	33,1	34,5	36,1	37,6	39,0	39,8	40,5	41,4	41,9
17	33,0	33,6	34,5	35,2	36,0	37,5	39,1	40,6	42,1	42,9	43,7	44,5	45,1
18	35,9	36,5	37,4	38,1	39,0	40,5	42,1	43,8	45,2	46,1	46,9	47,8	48,4
19	38,8	39,5	40,4	41,2	42,1	43,6	45,2	46,9	48,4	49,3	50,1	51,0	51,7
20	41,8	42,5	43,4	44,2	45,1	46,7	48,4	50,1	51,7	52,6	53,4	54,4	55,0
21	44,8	45,5	46,5	47,3	48,2	49,8	51,6	53,4	55,0	55,9	56,7	57,7	58,4
22	47,8	48,5	49,5	50,4	51,3	53,0	54,8	56,6	58,2	59,2	60,1	61,1	61,7
23	50,8	51,5	52,6	53,4	54,4	56,1	58,0	59,8	61,5	62,5	63,4	64,4	65,1
24	53,8	54,5	55,6	56,5	57,5	59,2	61,1	63,0	64,7	65,8	66,7	67,7	68,4
25	56,7	57,5	58,5	59,4	60,5	62,3	64,2	66,2	67,9	69,0	69,9	71,0	71,7
26	59,6	60,3	61,4	62,4	63,4	65,3	67,3	69,3	71,1	72,1	73,1	74,2	74,9
27	62,4	63,1	64,3	65,2	66,3	68,2	70,2	72,3	74,1	75,2	76,2	77,3	78,0
28	65,0	65,8	67,0	68,0	69,1	71,0	73,1	75,2	77,0	78,2	79,2	80,3	81,1
29	67,6	68,4	69,6	70,6	71,7	73,7	75,8	77,9	79,9	81,0	82,0	83,2	84,0
30	70,0	70,8	72,0	73,1	74,2	76,2	78,4	80,6	82,6	83,7	84,8	86,0	86,8
31	72,3	73,1	74,4	75,4	76,6	78,6	80,9	83,1	85,1	86,3	87,4	88,6	89,4
32	74,4	75,3	76,5	77,6	78,8	80,9	83,1	85,4	87,5	88,7	89,8	91,0	91,9
33	76,3	77,2	78,5	79,6	80,8	82,9	85,2	87,6	89,7	90,9	92,0	93,3	94,1
34	78,1	78,9	80,2	81,3	82,6	84,8	87,1	89,5	91,6	92,9	94,0	95,3	96,2
35	79,5	80,4	81,8	82,9	84,2	86,4	88,8	91,2	93,4	94,7	95,8	97,2	98,1
36	80,8	81,7	83,1	84,2	85,5	87,8	90,2	92,7	94,9	96,3	97,4	98,8	99,7
37	81,8	82,7	84,1	85,3	86,6	88,9	91,4	93,9	96,2	97,5	98,7	100,1	101,0
38	82,5	83,5	84,9	86,1	87,4	89,8	92,3	94,9	97,2	98,6	99,8	101,2	102,1
39	83,0	83,9	85,4	86,6	88,0	90,3	92,9	95,5	97,9	99,3	100,5	101,9	102,9
40	83,1	84,1	85,5	86,8	88,2	90,6	93,2	95,9	98,3	99,7	100,9	102,4	103,4
41	82,9	83,9	85,4	86,6	88,1	90,5	93,2	95,9	98,3	99,8	101,0	102,5	103,5

Table 2 Fitted centiles of head circumference (HC) (mm)

GA	0,5°	1°	2,5°	5°	10°	25°	50°	75°	90°	95°	97,5°	99°	99,5°
14	97	98	101	102	105	108	112	116	120	122	124	126	127
15	107	109	111	113	115	119	123	127	131	133	135	138	139
16	118	119	122	124	126	130	135	139	143	145	147	150	151
17	129	130	133	135	137	142	146	151	155	157	159	162	164
18	139	141	144	146	149	153	158	163	167	170	172	174	176
19	151	152	155	157	160	165	170	175	179	182	184	187	189
20	162	164	166	169	172	176	182	187	191	194	197	199	201
21	173	175	178	180	183	188	193	199	204	207	209	212	214
22	184	186	189	192	195	200	205	211	216	219	221	224	227
23	195	197	200	203	206	211	217	223	228	231	234	237	239
24	205	208	211	214	217	222	228	234	240	243	246	249	251
25	216	218	221	224	228	233	239	246	251	255	257	261	263
26	226	228	232	235	238	244	250	257	262	266	269	272	275
27	236	238	242	245	248	254	261	267	273	277	280	284	286
28	245	248	251	254	258	264	271	278	284	288	291	294	297
29	254	257	260	264	267	274	281	288	294	298	301	305	307
30	262	265	269	272	276	283	290	297	303	307	311	315	317
31	270	273	277	280	284	291	298	306	312	316	320	324	326
32	277	280	284	288	292	298	306	314	320	325	328	332	335
33	283	286	291	294	298	305	313	321	328	332	336	340	343
34	289	292	296	300	304	312	319	327	335	339	343	347	350
35	294	297	301	305	309	317	325	333	341	345	349	353	356
36	298	301	305	309	314	321	330	338	346	350	354	359	362
37	300	304	308	312	317	325	333	342	350	354	358	363	366
38	302	306	310	315	319	327	336	345	353	357	361	366	369
39	303	307	311	316	320	329	337	346	355	359	364	368	372
40	303	306	311	316	321	329	338	347	355	360	365	370	373
41	301	305	310	314	319	328	337	347	355	360	364	370	373

Table 3 Fitted centiles of abdominal circumference (AC) (mm)

GA	0,5°	1°	2,5°	5°	10°	25°	50°	75°	90°	95°	97,5°	99°	99,5°
14	76	78	80	82	84	88	92	96	100	102	104	106	108
15	86	87	90	92	94	99	103	108	112	114	116	119	120
16	95	97	100	102	105	109	114	119	123	126	128	131	133
17	105	107	110	112	115	120	125	130	135	138	140	143	145
18	115	117	120	123	126	131	136	142	147	150	153	156	158
19	125	127	130	133	136	142	148	154	159	162	165	168	170
20	135	137	140	143	147	152	159	165	171	174	177	181	183
21	144	147	150	154	157	163	170	177	183	186	189	193	196
22	154	157	161	164	168	174	181	188	194	198	202	205	208
23	164	166	170	174	178	185	192	199	206	210	214	218	220
24	173	176	180	184	188	195	203	211	218	222	226	230	233
25	183	186	190	194	198	206	214	222	229	234	238	242	245
26	192	195	200	204	208	216	224	233	241	245	249	254	257
27	201	204	209	213	218	226	235	244	252	257	261	266	269
28	210	213	218	223	228	236	245	254	263	268	272	277	281
29	218	222	227	232	237	246	255	265	274	279	283	289	292
30	227	231	236	241	246	255	265	275	284	289	294	300	303
31	235	239	245	249	255	264	275	285	294	300	305	310	314
32	243	247	253	258	264	273	284	295	304	310	315	321	325
33	251	255	261	266	272	282	293	304	314	320	325	331	335
34	258	262	268	274	280	290	302	313	323	330	335	341	345
35	265	269	276	281	287	298	310	322	332	339	344	351	355
36	271	276	282	288	295	306	318	330	341	348	353	360	364
37	277	282	289	295	301	313	325	338	349	356	362	369	373
38	283	288	295	301	308	320	332	345	357	364	370	377	382
39	288	293	301	307	314	326	339	352	364	371	378	385	390
40	293	298	306	312	319	332	345	359	371	379	385	392	397
41	297	303	310	317	324	337	351	365	378	385	392	399	405

Table 4 Fitted centiles of femur length (FL) (mm)

GA	0,5°	1°	2,5°	5°	10°	25°	50°	75°	90°	95°	97,5°	99°	99,5°
14	11,6	12,0	12,5	13,0	13,6	14,5	15,5	16,6	17,5	18,1	18,5	19,1	19,5
15	14,6	14,9	15,5	16,0	16,6	17,6	18,7	19,7	20,7	21,3	21,8	22,4	22,8
16	17,5	17,9	18,5	19,0	19,6	20,6	21,7	22,8	23,8	24,4	25,0	25,6	26,0
17	20,3	20,7	21,4	21,9	22,5	23,6	24,7	25,9	26,9	27,5	28,1	28,7	29,1
18	23,1	23,5	24,2	24,8	25,4	26,5	27,7	28,9	29,9	30,6	31,1	31,8	32,2
19	25,8	26,3	27,0	27,5	28,2	29,3	30,6	31,8	32,9	33,6	34,2	34,8	35,3
20	28,5	29,0	29,7	30,3	31,0	32,1	33,4	34,7	35,8	36,5	37,1	37,8	38,3
21	31,1	31,6	32,3	33,0	33,7	34,9	36,2	37,5	38,7	39,4	40,0	40,7	41,2
22	33,7	34,2	34,9	35,6	36,3	37,5	38,9	40,3	41,5	42,2	42,8	43,6	44,1
23	36,2	36,7	37,5	38,1	38,9	40,1	41,5	42,9	44,2	45,0	45,6	46,4	46,9
24	38,6	39,1	39,9	40,6	41,4	42,7	44,1	45,6	46,9	47,6	48,3	49,1	49,6
25	41,0	41,5	42,3	43,0	43,8	45,1	46,6	48,1	49,5	50,3	50,9	51,8	52,3
26	43,2	43,8	44,6	45,4	46,2	47,5	49,1	50,6	52,0	52,8	53,5	54,3	54,9
27	45,4	46,0	46,9	47,6	48,5	49,9	51,4	53,0	54,4	55,3	56,0	56,9	57,4
28	47,6	48,2	49,1	49,8	50,7	52,1	53,7	55,3	56,8	57,7	58,4	59,3	59,9
29	49,6	50,2	51,2	51,9	52,8	54,3	55,9	57,6	59,1	60,0	60,7	61,6	62,3
30	51,6	52,2	53,2	54,0	54,9	56,4	58,1	59,8	61,3	62,2	63,0	63,9	64,5
31	53,5	54,2	55,1	55,9	56,8	58,4	60,1	61,9	63,4	64,4	65,2	66,1	66,8
32	55,3	56,0	57,0	57,8	58,7	60,3	62,1	63,9	65,5	66,4	67,3	68,2	68,9
33	57,0	57,7	58,7	59,6	60,5	62,2	64,0	65,8	67,4	68,4	69,3	70,2	70,9
34	58,7	59,4	60,4	61,3	62,3	63,9	65,8	67,6	69,3	70,3	71,2	72,2	72,9
35	60,2	60,9	62,0	62,9	63,9	65,6	67,5	69,4	71,1	72,1	73,0	74,0	74,7
36	61,7	62,4	63,5	64,4	65,4	67,2	69,1	71,0	72,8	73,8	74,7	75,8	76,5
37	63,0	63,8	64,9	65,8	66,8	68,6	70,6	72,6	74,4	75,4	76,4	77,4	78,2
38	64,3	65,1	66,2	67,1	68,2	70,0	72,0	74,0	75,9	77,0	77,9	79,0	79,8
39	65,5	66,2	67,4	68,3	69,4	71,3	73,4	75,4	77,3	78,4	79,3	80,5	81,2
40	66,5	67,3	68,5	69,4	70,6	72,5	74,6	76,7	78,6	79,7	80,7	81,8	82,6
41	67,5	68,3	69,5	70,5	71,6	73,5	75,7	77,8	79,8	80,9	81,9	83,1	83,9

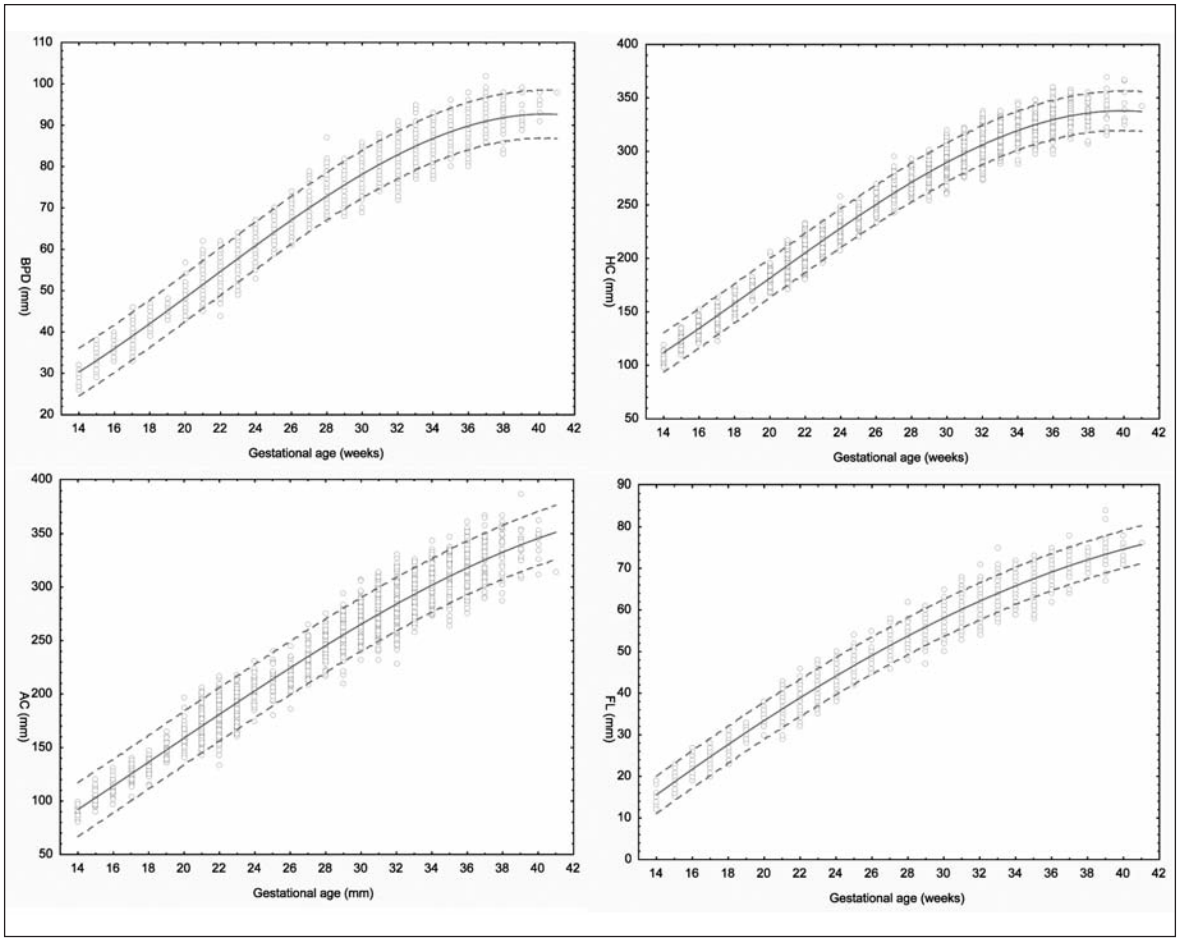

Figure 1 Fitted 5th, 50th and 95th centile curves superimposed on the raw data for biparietal diameter (BPD), head circumference (HC), abdominal circumference (AC) and femur length (FL). Dashed lines represent the 5th and 95th centiles and the thick central one the 50th centile.

The representation of the fetal equations into Z-scores, as suggested by Salomon, lead to evaluate the effect of the reference curve chosen on the quality of screening for growth abnormalities. Indeed, knowing only the mean and the standard deviation, the Z-score allows a comparison between different curves. In our series, the graphical representation highlights that Italian fetuses have smaller HC and AC than English and French fetuses. This may suggest that Italian fetuses are smaller than other populations or that it may arise from the different methods used by Salomon in the measurement of HC and AC, which were manually calculated from BPD and FOD and the two maximum abdominal diameters, respectively. Finally Italian fetuses seem to have much smaller femurs than the English, at least until the beginning of the third trimester, but similar length than French femurs.

Conclusion

Our new Italian reference interval charts for fetal biometry and reference equations have clinical relevance since they provide sonographers new reference equations in the obstetrical practice.

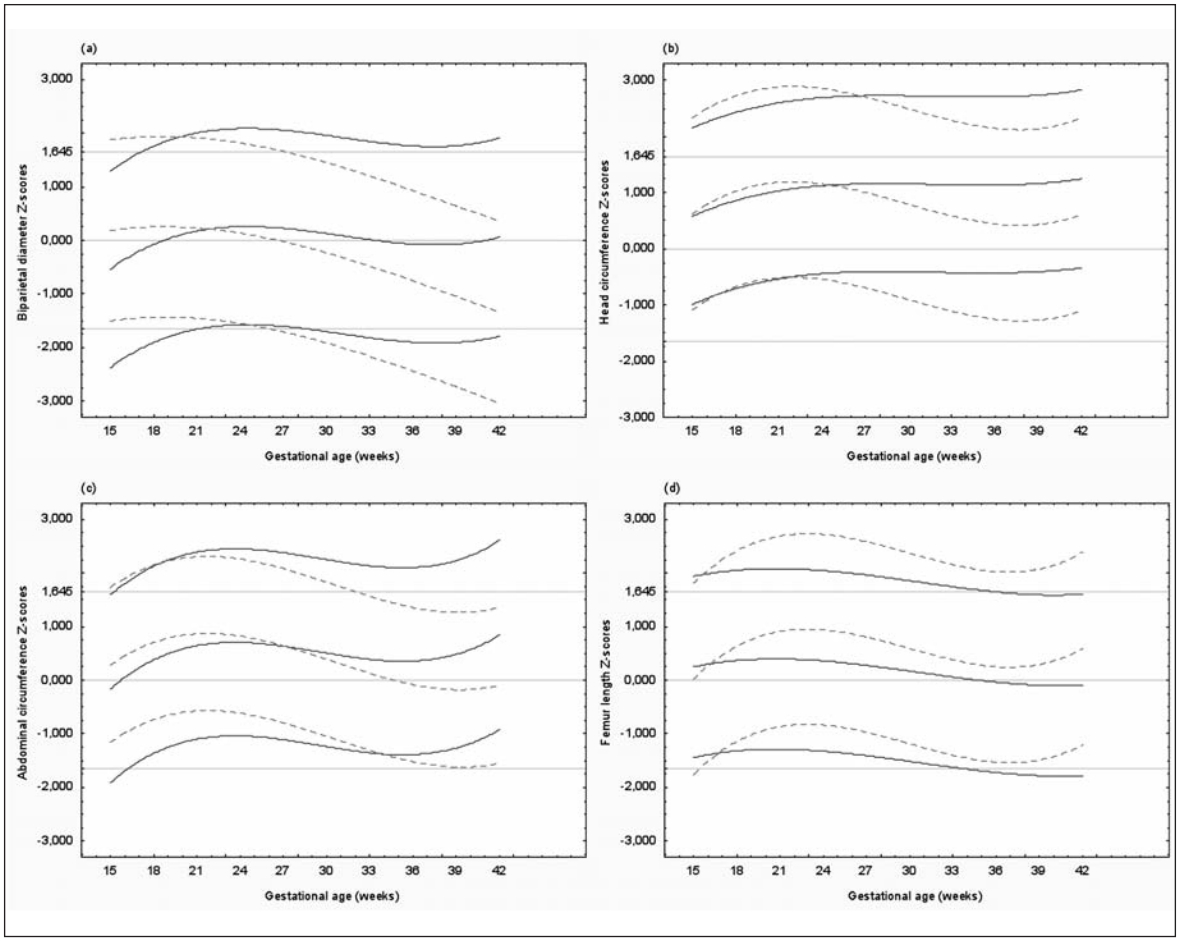

Figure 2 Comparison of our new equations with Salomon (solid lines) and Snijders (dashed lines) references for biparietal diameter (a), head circumference (b), abdominal circumference (c) and femur length (d). Dotted gridlines represent the expected Z-scores for 5th, 50th and 95th centiles, i.e. -1.645, 0 and 1.645 respectively.

References

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