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Poor ovarian response as a predictor for live birth in older women undergoing IVF

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KEY MESSAGE

Poor response to stimulation in the first IVF cycle has a limited predictive value for the outcomes of subsequent cycles in women over 40 years. Poor response, defined by number of oocytes retrieved, and not by number of dominant follicles, is an independent predictor for births for women over 40.

A B S T R A C T

Women of advanced age present a major challenge for fertility treatments. This study was designed to assess whether poor ovarian response (POR) according to the Bologna criteria is a significant predictor for live birth in women over 40. The outcomes of subsequent IVF cycles were also studied. The results of 1870 fresh IVF cycles in 1212 women were retrospectively analysed. The live birth per cycle was 3.3 times higher (11.61% versus 3.54%, $P < 0.001$) in good responders with more than three oocytes collected compared with women with less. Ovarian response defined by oocytes collected, but not by the number of follicles, was independently associated with live birth (odds ratio, 2.0; 95% confidence interval, 1.18 to 3.54; $P = 0.009$). The occurrence of POR in subsequent IVF cycles was only 55%. No differences in live births were found in persistent POR compared with women with at least one good response. A single episode of POR in a first IVF cycle in older women has a limited predictive value for the outcomes of subsequent cycles. POR in women aged 40–43 years, defined by the number of oocytes retrieved, is a predictor for live birth in IVF.

Introduction

As more women delay childbearing, women at the age of 40 and over present a major challenge for fertility treatments. Spontaneous live birth rate decreases dramatically in this age group and, even with IVF, the live birth rate for this subgroup of patients is around 10% (Crawford et al., 2017; Sunderam et al., 2017). Earlier single-centre studies reported a live birth rate of 2.0–13.9% for women between the ages of 40 and 43 (Klipstein

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et al., 2005; Lass et al., 1998; Serour et al., 2010; Tsafrir et al., 2007).

The decline in fertility in older women over 40 is caused both by reduced ovarian reserve caused by accelerated follicular loss (Faddy et al., 1992), as well as by a higher rate of chromosomal abnormalities and cytoplasmic dysfunction, such as mitochondrial dysfunction (Hassold and Hunt, 2009; Miao et al., 2009). These destructive mechanisms will ultimately lead to lower pregnancy rates and higher pregnancy losses. Poor ovarian response (POR) to ovarian stimulation is defined either by ovarian reserve tests such as Day 3 FSH, antral follicle count (AFC), the number of growing follicles during controlled ovarian stimulation or, alternatively, following ovum pickup, by the number of oocytes retrieved. Since 2011, the accepted definition for women with POR is the Bologna criteria (Ferraretti et al., 2011).

The success rate of IVF is correlated with the ovarian response and the number of oocytes retrieved (Sunkara et al., 2011). Nevertheless, few studies have evaluated the outcome of IVF in women over 40 according to their ovarian response. Biljan et al. (2000) used sonographic criteria to define poor response and indicated a higher but not statistically significant live birth rate per cycle in those with more than three follicles (15.7% versus 4.2%). Yih et al. (2005) found a higher clinical pregnancy rate in women above 40 when five or more oocytes were retrieved. The Bologna criteria for poor responders (Ferraretti et al., 2011) standardizes the definition of POR. Women aged 40 and over need only one additional criteria to meet the definition of POR, including either previous POR (≤3 oocytes with conventional stimulation) or an abnormal ovarian test reserve.

Although one may assume that older women with good response carry a better prognosis for live birth, the effect of lower oocyte quality cannot be ignored. The aim of this study was to assess ovarian response based on the number of oocytes retrieved in accordance with the Bologna criteria and the number of dominant follicles, as significant predictors for live birth rate in women aged 40–43.

Materials and methods

Patient population

This was a retrospective study conducted at a university hospital reproductive centre. The computerized database of the IVF unit for the years 2010 to 2015 was retrospectively analysed. The analysis included women who were 40–43 years of age, undergoing a non-donor IVF cycle. Pre-implantation genetic diagnosis (PGD), oocyte donation, in-vitro maturation (IVM) and natural IVF cycles were excluded. Women older than 43 were not treated, according to clinic policy. The study received Institutional Review Board approval on 16 May 2013 (reference number 13–053-SDR).

IVF was publicly funded during the study period. According to the clinic policy, all patients with at least one follicle underwent ovum pickup. The outcome measures were calculated for women with at least one oocyte collected (per oocyte pickup). We analysed the primary outcome measures according to two accepted definitions for POR to stimulation: the first complies with the Bologna criteria – ≤ 3 oocytes retrieved and age 40 and above (Ferraretti et al., 2011); the second is based on ultrasonographic findings of ≤3 dominant follicles (>14 mm in diameter) imaged 24–48 h prior to human chorionic gonadotrophin (HCG) administration (potential POR) (Biljan et al., 2000; Fridström et al., 1997).

The primary outcome measure was live birth. Secondary outcome measures included IVF protocol parameters, embryo development and baseline ovarian reserve test results. A second analysis was conducted for women over 40 undergoing their first IVF cycle. In addition, we searched for the outcome of poor responders in their first cycle with one or more subsequent IVF cycles. Those who had ≤3 oocytes in all subsequent cycles were defined as persistent poor responders and were compared with women with at least one subsequent cycle with good response. The outcome of women with cryopreserved embryos that underwent cryopreserved embryo transfer was also analysed.

Ovarian stimulation protocol

Ovarian stimulation was performed by means of one of the following protocols: microdose-flare protocol, fixed gonadotrophin-releasing hormone (GnRH) antagonist protocol and mid-luteal long GnRH agonist protocol. Briefly, ovarian stimulation was performed with gonadotrophins (Gonal-F, EMD-Serono Inc., Canada; Puregon, Merck Inc., Canada; Menopur, Ferring Pharmaceuticals Inc., Canada), ovulation was suppressed with GnRH antagonist (Cetrotide, EMD-Serono Inc.; Orgalutran, Merck Inc.) or GnRH agonist (Superfact, Sanofi-Aventis Inc., Canada). No standard stimulation protocol is used for patients with POR in our clinic; protocol and gonadotrophin dose were based on AFC, Day 3 FSH and the results of previous IVF cycles. Gonadotrophin dose was ≥300 IU FSH per day. After 5–7 days of stimulation, FSH dose was adjusted based on ultrasound scan and serum oestradiol level. The maximal dose of FSH was 600 IU. When ideally two follicles attained a mean diameter of 17 mm, 250 µg of human recombinant chorionic gonadotrophin was administered (Ovidrel, EMD-Serono Inc.). Oocyte retrieval by an ultrasound guided transvaginal approach was scheduled 36–38 h later.

Embryo culture

Oocytes were fertilized by either conventional IVF or intracytoplasmic sperm injection (ICSI), according to the clinical indication. Fertilization was assessed 16–18 h after insemination for the appearance of two distinct pronuclei and two polar bodies. The zygotes were cultured in cleavage medium (Cook Medical, Sydney, Australia). Embryonic development was assessed daily. Embryos were cultured to the blastocyst stage in the blastocyst medium (Cook Medical). According to our IVF laboratory protocol, if there were at least three good-quality embryos on Day 3, the embryo culture was extended to the blastocyst stage and the embryos were transferred on Day 5. Cleavage embryos were defined as good quality (Grade 1 or 2) if they had four cells on Day 2 and/or seven or eight cells on Day 3, contained <20% fragmentation and exhibited no apparent morphological abnormalities. Poor-quality embryos included fair quality (Grade 3) embryos, which had only two cells on Day 2, three to five cells on Day 3 and/or 20–50% fragmentation, and Grade 4 embryos with <3 cells by Day 3 and >50% fragmentation. For women with fewer than three good-quality embryos, embryo transfer was at Day 2–3. Supernumerary good-quality embryos were cryopreserved (Zhang et al., 2011). The number of embryos transferred was determined according to the provincial guidelines at that time: up to two or three embryos and never more than three.

Outcome measures

The primary outcome was live birth rate per oocyte pickup. Live birth was defined as delivery of a live fetus ≥24 weeks of pregnancy. A

pregnancy was defined by a positive serum β-HCG test 11–14 days after embryo transfer and clinical pregnancy was defined by ultrasound visualization of an intrauterine gestational sac with fetal heartbeat.

Statistical analysis

The data were not normally distributed, and so a non-parametric Wilcoxon signed-rank test was used to assess differences in population mean ranks of continuous variables. Categorical variables were compared by chi-squared test or Fisher's exact test, when appropriate. Descriptive statistics are given as median (interquartile range, IQR) for skewed data. Moreover, in order to examine which parameters are associated with live births, a stepwise multiple logistic regression model, adjusted for age, number of embryos transferred, availability of embryos for cryopreservation, Day 3 FSH, AFC, undergoing subsequent IVF cycles and good response defined by oocytes or by dominant follicles, was applied. A *P*-value <0.05 was considered significant. All statistical analysis was performed using JMP 12 for Macintosh (SAS Institute Inc., Cary, NC, USA).

Results

The medical records of 1278 women at the age of 40–43 with 1941 fresh IVF cycles were retrieved. The analysis included the outcomes of 1870 fresh IVF cycles (1212 women) with at least one oocyte retrieved. Poor response was documented in 509 cycles with ≤3 oocytes retrieved and in 851 cycles with ≤3 dominant follicles visualized ultrasonographically; of these, in 39 cycles only one dominant follicle was visualized. The average number of cycles per patient was 1.92 ± 0.9 (the number of women undergoing one, two, three and more than three cycles was 720, 667, 414 and 69, respectively). A total of 361 women had been pregnant before undergoing IVF and 360 had given birth (mean gravidity and parity were 0.6 and 0.34, respectively). Similar proportions of women with at least one previous pregnancy or live birth were found in women defined as good as compared with poor responders. Additional aetiology of infertility, except for patient age, was documented in 913 women: unexplained infertility (30%), decreased ovarian reserve (24%), male factor (24%), tubal factor (6%), ovulatory dysfunction (4%), multiple factors (9%) and endometriosis (3%) (**Supplementary Table S1**).

The overall clinical pregnancy rate was 15.0% and the live birth rate was 9.4%. Clinical pregnancy and live birth rates analysed separately for women in their first IVF cycle were similar: 15.4% and 9.0%, respectively. Mean gestational age at delivery was 38 weeks (range 27–42 weeks), mean birth weight was 3188 g (range: 1060–4850 g). Ten women delivered twins at average week of delivery 36 (range 31–39 weeks) and 36.92% of clinical pregnancies spontaneously miscarried. Live birth per year of age is presented in **Figure 1**. In total, 204 women underwent subsequent cryopreserved embryo transfer, resulting in 17 live births (8.3%). Of these transfers only five were in women with POR, as defined by the number of oocytes retrieved (≤3), and these resulted in the birth of one baby.

In this cohort, cycles were not cancelled as a result of POR and all patients with at least one follicle underwent ovum pickup. However, of the cycles in which at least one oocyte was retrieved, 199 cycles did not reach embryo transfer, 116 due to failed fertilization, 24 due to very poor embryo development of non-viable embryos, 16 due to the risk of ovarian hyperstimulation syndrome, 9 due to uterine factors (polyps, thin endometrium) and 34 in which sperm could not be retrieved. In 71 cycles (3.7%) no oocytes were retrieved.

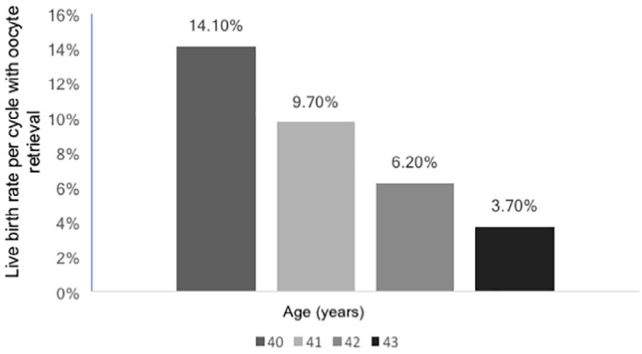

Figure 1 – Live birth rate according to age.

The differences in patient characteristics and protocol parameters between women with good response versus poor response are presented in **Table 1**. It is worth noting that women with a good response to ovarian stimulation, defined either by the number of oocytes or the number of dominant follicles, had a higher number of cleavage stage embryos, cycles with embryos available for cryopreservation and cycles with blastocyst transfers. The live birth per oocyte retrieval was 3.3 times higher (11.61% versus 3.54%, $P < 0.001$) in good responders with >3 oocytes collected, compared with 1.8 times higher (11.78% versus 6.58%, $P < 0.001$) in women with >3 dominant follicles when compared with women with fewer (**Table 1**).

Similar differences in protocol parameters and cycle outcome were found in a subgroup analysis for first IVF cycle only. Higher live birth rates were recorded in good responders, whether this was defined by oocytes collected or dominant follicles visualized ultrasonographically (**Table 2**). More cycles in poor responders did not attain embryo transfer due to failed fertilization and very poor embryo development.

Outcomes of 353 women undergoing their first IVF cycle who did not achieve pregnancy and who had one or more subsequent cycles were analysed. Respectively, 225, 115 and 13 women had one, two and three subsequent IVF trails. Forty per cent (37/93) of women who responded poorly in their first IVF cycle, as defined by the number of oocytes collected, were good responders in the second cycle. Of these 37, 10 had no change in their stimulation protocol, 5 had an increased gonadotrophin dose and 22 had a different stimulation protocol. Nine per cent (5/54) from those who responded poorly in their first two cycles were good responders in the third cycle. Fifty-one women (54.8%) were persistent poor responders. Live birth rate per patient was comparable in persistent poor responders and women with at least one subsequent cycle with good ovarian response (9.8% [5/51] versus 11.9% [5/42]). A similar live birth rate of 8.7% (20/229) was recorded in good responders in their first cycle. Live birth rates for poor responders defined by the number of oocytes were 3.3% (7/211), 8.8% (8/91), 6.9% (2/29) and 0% (0/1) for the first, second, third and fourth IVF cycle, respectively.

The association between live birth and age, availability of embryos for cryopreservation, number of cryopreserved embryos, AFC, Day 3 FSH, undergoing subsequent IVF cycles, good response defined by oocytes or number of dominant follicles, was analysed using a stepwise logistic regression analysis (**Table 3**). In the analysis of all IVF cycles, ovarian response defined by >3 oocytes collected (odds ratio

Table 1 – Differences in patient characteristics, protocol parameters and IVF outcome between women with good response versus poor response.

	Good response >3 oocytes	Poor response ≤3 oocytes	*P*-value	Good response >3 DF	Poor response ≤3 DF	*P*-value
n	1361	509		1019	851	
Age (years)	41.13 ± 0.98	41.37 ± 0.96	<0.001	41.14 ± 0.97	41.26 ± 0.98	0.008
Baseline ovarian reserve tests						
Day 3 FSH (IU/l)	7.4 (6.0–8.8)	8.2 (6.9–11.8)	<0.001	7.2 (5.9–8.6)	8.2 (6.75–10.4)	<0.001
AFC	12 (7–17)	6 (4–8)	<0.001	12 (8–18)	7 (5–11)	<0.001
Protocol						
MDF, *n* (%)	698 (51.3)	305 (59.9)	0.001	554 (54.4)	451 (53.0)	NS
Long agonist, *n* (%)	310 (22.8)	75 (14.7)	<0.001	199 (19.5)	185 (21.7)	NS
Antagonist, *n* (%)	352 (25.9)	129 (25.3)	NS	266 (26.1)	215 (25.3)	NS
IVF protocol parameters						
Total FSH (IU)	3600 (2358–4875)	4500 (3150–5850)	<0.001	3450 (2250–4950)	4050 (3000–5400)	<0.001
FSH (IU) per oocyte	406 (232–720)	2100 (1462–3600)	<0.001	366 (212–693)	1200 (600–2250)	<0.001
Peak serum oestradiol (pmol/l)	6300 (4539–9005)	2482 (1626–3689)	<0.001	6620 (4989–12255)	3261 (2253–5079)	<0.001
DF >14 mm	5 (3–7)	2 (1–3)	<0.001	6 (5–8)	2 (1–3)	<0.001
Oocytes collected	7 (5–11)	2 (2–3)	<0.001	8 (6–12)	3 (2–5)	<0.001
MII oocytes	5 (4–8.5)	2 (1–2)	<0.001	6 (4–9)	3 (2–4)	<0.001
ICSI, *n* (%)	1095 (80.46)	406 (79.76)	NS	821 (80.57)	680 (79.91)	NS
Two pronuclei (2PN)	4 (2–6)	1 (1–2)	<0.001	4 (2–7)	2 (1–3)	<0.001
No. of cleavage stage embryos	4 (2–6)	1 (1–2)	<0.001	4 (2–6)	2 (1–3)	<0.001
Embryo grade[a]						
Grade 1 or 2, *n* (%)	627/888 (70.6)	239/391 (61.1)	0.002	453/629 (72)	414/650 (63.7)	0.004
Grade 3, *n* (%)	256/888 (28.8)	141/391 (36.1)	0.018	171/629 (27.2)	224/650 (34.5)	0.009
Grade 4, *n* (%)	4/888 (0.5)	11/391 (2.8)	0.003	4/629 (0.6)	10/650 (1.5)	NS
Embryo cryopreservation[b], *n* (%)	286 (21.01)	5 (0.98)	<0.001	244 (23.95)	47 (5.52)	<0.001
Cycles with blastocyst transfer, *n* (%)	385 (28.3)	7 (1.4)	<0.001	319 (31.3)	73 (8.6)	<0.001
No. of embryos transferred	2 (1–2)	1 (1–2)	<0.001	2 (1–2)	1 (1–2)	<0.001
IVF cycle outcomes						
No embryo transfer, *n* (%)	88 (6.47)	111 (21.81)	<0.001	71 (6.97)	128 (15.04)	<0.001
Biochemical pregnancy, *n* (%)	336 (24.69)	51 (10)	<0.001	257 (25.22)	130 (15.28)	<0.001
Clinical pregnancy, *n* (%)	247 (18.1)	32 (6.3)	<0.001	185 (18.16)	94 (11.05)	<0.001
Live birth, *n* (%)	158 (11.61)	18 (3.54)	<0.001	120 (11.78)	56 (6.58)	<0.001
Miscarriage, *n* (%)	89/247 (36)	14/32 (43.75)	NS	65/185 (35.14)	38/94 (40.43)	NS

Data presented as mean ± standard deviation or median (interquartile range).
AFC = antral follicle count; DF = dominant follicle; MDF = micro-dose flare; MII = metaphase II; NS = not significant.
[a] Grade of the best cleavage stage embryo transferred. Good quality: grade 1 or 2, fair quality: grade 3, poor quality: grade 4.
[b] Number of cycles with embryo cryopreservation.

[OR], 2.0; 95% confidence interval [CI], 1.18 to 3.54), but not by the number of mature follicles, was associated with live birth. The following factors were also found to be significantly associated with live birth: age (OR, 0.65; 95% CI, 0.54 to 0.78), number of embryos transferred (OR, 1.83; 95% CI, 1.42 to 2.37), cycles with cryopreserved embryos (OR, 2.6; 95% CI, 1.78 to 3.81) and women undergoing more than one IVF cycle (OR, 0.26; 95% CI, 0.19 to 0.37). In a subgroup analysis of women undergoing their first cycle: age (OR, 0.64; 95% CI, 0.49 to 0.85), number of embryos transferred (OR, 1.56; 95% CI, 1.06 to 2.29) and embryos available for cryopreservation (OR, 2.64; 95% CI, 1.5 to 4.64) were significant predictors for live birth rate.

Discussion

In general, women with good ovarian response during IVF would have more oocytes, more embryos and a higher chance of pregnancy and birth. Nevertheless, with age, both oocyte quantity and quality are compromised (Faddy et al., 1992; Hassold and Hunt, 2009). To what extent does poor oocyte quality in this subgroup of IVF patients affect the live birth rate? In this study, we found that the live birth rate is up to 3.3 times higher in older women who had good responses, defined by >3 oocytes collected. Alternatively, if ovarian response is estimated by the number of dominant follicles visualized ultrasonographically, the live birth rate is 1.8 times higher in women with >3 dominant follicles. Similar live birth rates were found when analysing women undergoing their first IVF cycle.

A single episode of POR to stimulation in the first IVF cycle has a limited predictive value for the outcomes of subsequent cycles. The results of this study do not support using it to recommend against further cycles. Fifty-five per cent of women with ≤3 oocytes in their first IVF cycle will respond poorly to ovarian stimulation in subsequent cycles. This is in accordance with previous studies which found that the recurrence rate of poor response was 54–62% (Klinkert et al., 2004; Peñarrubia et al., 2005). Remarkably, it appears that the rate of persistent poor responders is not affected by the older age of participants in this study. Furthermore, comparable live birth rates in persistent poor responders versus women with at least one cycle with good response also does not support withdrawal from further IVF attempts in women who responded poorly in their first IVF cycle. The live birth rates in the second and third IVF cycle in women who were

Table 2 – Differences in patient characteristics, protocol parameters and outcome in the first IVF cycle between women with good response versus poor response.

	Good response >3 oocytes	Poor response ≤3 oocytes	*P*-value	Good response >3 DF	Poor response ≤3 DF	*P*-value
n	561	211		411	360	
Age (years)	41.07 ± 0.98	41.33 ± 0.99	0.001	41.09 ± 0.98	41.19 ± 1.00	NS
Baseline ovarian reserve tests						
Day 3 FSH (IU/l)	7.3 (5.9–8.8)	8.4 (7.45–16.45)	0.001	7.2 (5.9–8.55)	8.3 (6.68–11.33)	0.002
AFC	11 (7–17)	5.5 (2–8.75)	<0.001	12 (8–17)	7 (5–13)	<0.001
IVF protocol parameters						
Total FSH (IU)	3000 (2025–4500)	4050 (3000–5737)	<0.001	3000 (2025–4800)	3600 (2700–4950)	<0.001
FSH units per oocyte (IU)	366 (197–652)	2200 (1350–3600)	<0.001	350 (180–616)	1012 (540–2250)	<0.001
Peak serum oestradiol (pmol/l)	6420 (4724–9403)	2379 (1512–3371)	<0.001	6689 (5081–9930)	3317 (2381–5244)	<0.001
DF >14 mm	5 (3–7)	2 (1–3)	<0.001	6 (5–8)	2 (2–3)	<0.001
Oocytes collected	7 (5–11)	2 (1–3)	<0.001	8 (5–12)	4 (2–6)	<0.001
MII oocytes	5 (4–9)	2 (1–2)	<0.001	6 (4–10)	3 (2–4)	<0.001
ICSI, *n* (%)	391 (69.7)	152 (72.04)	NS	286 (69.6)	257 (71.39)	NS
Two pronuclei (2PN)	4 (2–6)	1 (1–2)	<0.001	4 (2–7)	2 (1–3)	<0.001
No. of cleavage stage embryos	4 (2–6)	1 (1–2)	<0.001	4 (2–7)	2 (1–3)	<0.001
Embryo cryopreservation[a], *n* (%)	143 (25.49)	2 (0.95)	<0.001	120 (29.2)	24 (6.67)	<0.001
Cycles with blastocyst transfer, *n* (%)	183 (32.6)	2 (0.95)	<0.001	152 (37)	34 (9.4)	<0.001
No. of embryos transferred	2 (1–2)	1 (1–2)	<0.001	2 (1–2)	1 (1–2)	0.02
IVF cycle outcomes						
No embryo transfer, *n* (%)	31 (5.53)	41 (19.43)	<0.001	27 (6.57)	45 (12.50)	0.005
Biochemical pregnancy, *n* (%)	144 (25.67)	20 (9.48)	<0.001	103 (25.06)	61 (16.94)	0.006
Clinical pregnancy, *n* (%)	106 (18.89)	13 (6.16)	<0.001	72 (17.52)	47 (13.06)	NS
Live birth, *n* (%)	69 (12.30)	7 (3.32)	<0.001	51 (12.41)	25 (6.94)	0.01
Miscarriage, *n* (%)	37/106 (34.9)	6/13 (46.15)	NS	21/72 (29.17)	22/47 (46.8)	NS

Data presented as mean ± standard deviation or median (interquartile range).
AFC = antral follicle count; DF = dominant follicle; MII = metaphase II; NS = not significant.
[a] Number of cycles with embryo cryopreservation.

Table 3 – Odds ratio and 95% confidence intervals for predictors of live birth in women over 40 years undergoing IVF.

	Adjusted odds ratio	95% confidence interval	*P*-value
For all IVF cycles			
>3 oocytes retrieved	2.0	1.18–3.54	0.009
Age	0.65	0.54–0.78[a]	<0.001
Number of embryos transferred	1.83	1.42–2.37[a]	<0.001
Availability of embryos to cryopreserve	2.60	1.78–3.81	<0.001
Undergoing more than one IVF cycle	0.26	0.19–0.37	<0.001
For first cycle only			
Age	0.64	0.49–0.85[a]	0.001
Number of embryos transferred	1.56	1.06–2.29[a]	0.021
Availability of embryos to cryopreserve	2.64	1.5–4.64	<0.001
>3 oocytes retrieved	2.21	0.94–5.22	NS

Stepwise logistic regression to analyse the association between live birth and age, number of embryos transferred, availability of embryos for cryopreservation, antral follicle count, Day 3 FSH, undergoing subsequent IVF cycles, good response defined by oocytes or number of dominant follicles.
[a] Per unit change.
NS = not significant.

persistent poor responders was 7–8% per cycle with oocyte pickup. This result further supports our conclusion that poor response in women over 40 undergoing their first cycle is not a good indicator for the outcome of subsequent IVF trials.

As expected, IVF protocol parameters were related to ovarian response. Good responders required less total FSH dose and the FSH per oocyte was also significantly lower. A lower birth rate in poor responders was the result of a higher number of cycles without embryo transfer, fewer embryos available for transfer, lower embryo quality and fewer blastocyst stage transfers. Nevertheless, poor response cannot predict a miscarriage as the miscarriage rate was similar between all groups. Good response defined by >3 oocytes collected, but not according to the number of dominant follicles visualized ultrasonographically, was found to be an independent predictor for live birth.

The cohort of women that responded poorly to ovarian stimulation had several negative pretreatment prognostic factors. The prevalence of unexplained, male factor, ovulatory dysfunction infertility was lower, while decreased ovarian reserve and multifactor infertility were significantly higher in poor responders. Furthermore, the more favourable prognosis of good responders is reflected in their ovarian reserve tests. The differences in ovarian reserve tests were more prominent when analysed according to the AFC, although the differences in Day 3 FSH were also statistically significant. AMH and AFC have limited accuracy and a high false positive rate of 10–20% for predicting ovarian reserve (Broekmans et al., 2006; La Marca et al., 2010). They were found to correlate with the response

to ovarian stimulation, but were poor predictors for pregnancy and the live birth rate (Fauser *et al.*, 2007). For that reason, the ESHRE consensus suggests using both the women's age as well as ovarian response to stimulation as post hoc tests to confirm POR (Ferraretti et al., 2011).

The live birth rate reported in this study is in accordance with previous studies that reported a birth rate of 4.0–7.6% at 43 years of age and 9.0–13.9% at 40 years of age (Klipstein et al., 2005; Tsafrir et al., 2007). Moreover, the largest published single-centre study was conducted in 2003–2008 and indicated a live birth rate per oocyte pickup of 2.4% for 43 years to 11% at 40 years of age. The live birth rates stratified by age were comparable with our results. Sunkara et al. (2011) found a live birth rate of 1–6% for women over 40 with one to three oocytes collected, respectively. Poor responders were not compared with good responders per se and differences in protocol parameters and other IVF outcomes were not reported in that large national database analysis. Only very few studies analysed the effect of ovarian response to stimulation on the live birth rate using different methodology and cut-off values for poor response. Most importantly, they were published before the ESHRE consensus on the definition of POR, thus they did not use the accepted cut-off values. Biljan et al. (2000) reported a higher live birth rate in women over 40 with more than three follicles. However, due to the small sample size, it lacked statistical power. Ciray et al. (2006) found a higher delivery rate in women 40 years of age and over for those with more than six oocytes retrieved (15% versus 5.9%). Tsafrir et al. (2007) found higher pregnancy rates for women with more than five oocytes collected; however, the live birth rate was not reported. Similarly, Yih et al. (2005) also found significantly lower pregnancy rates in women with ≤4 oocytes retrieved, but only in women with normal ovarian reserve tests. Because the miscarriage rate in that age group is very high, reports without live birth figures give us limited prognostic value for the ultimate outcome of IVF.

The results of our multivariate analysis are in accordance with those of Polyzos et al. (2014), who found that the number of oocytes retrieved was a significant predictor associated with live birth in poor responders fulfilling the Bologna criteria across a wide range of ages. Our results confirm their findings in women over 40 years of age. However, other parameters like age, the number of embryos transferred and the availability of embryos for cryopreservation were also found to be associated with live birth. These differences can be explained by the larger sample size of our study, inclusion only of women over 40 and different adjustment for confounding factors in the logistic regression.

The results of this study show that the number of oocytes collected, and not the number of dominant follicles visualized ultrasonographically, is an independent predictor for live birth in women over 40. It could be explained by variability in follicle measurements by ultrasound. Moreover, oocytes could not be retrieved from some follicles during oocyte retrieval. Thus POR, as defined by the Bologna criteria, according to the number of oocytes retrieved, is a better marker for successful IVF cycles.

This is not the first study analysing IVF outcome in this age group. Nevertheless, most of the previous reports were from 1995 to 2008, thus this study represents the results of a modern IVF unit. This study also contributes to the limited available data on the success rate of women who responded poorly in their first IVF cycle who underwent further cycles. Because cycles were not cancelled prior to ovum pickup due to poor response, this analysis provides a good estimate of the success rate for this subgroup of patients, without the risk of bias caused by cycles cancelled due to poor response. Furthermore, although the predictive power of the number of oocytes was reported, comparison of poor responders versus good responders was not done according to the Bologna criteria. In addition, we analysed the data according to the number of dominant follicles for the added value of predicating the success rate of the cycle before ovum pickup. This study is limited by its retrospective design. AMH values could not be analysed as they are not part of the provincial routine tests, although it was suggested that AFC is a reliable marker to predict ovarian response to stimulation (Hsu et al., 2011).

In conclusion, poor response to ovarian stimulation in women over 40 years of age, as defined by the number of oocytes retrieved and not by the number of dominant follicles visualized ultrasonographically, is an independent predictor for live birth in IVF. POR in a first IVF cycle cannot be used to predict the recurrence of poor response in subsequent cycles. These women should be informed of their prognosis.

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Appendix: Supplementary material

Supplementary data to this article can be found online at doi:10.1016/j.rbmo.2018.01.008.

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