Diminished ovarian reserve and adverse reproductive outcomes

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Chapter 7

Summary, general discussion and future perspectives
The aim of this thesis is to determine whether there is an association between diminished ovarian reserve and adverse reproductive outcomes or reproductive lifespan (menopause). The association between quantity and quality of oocytes is important to be established since women are trying to conceive at a later age, when the ovarian reserve is diminished to a level in which there are higher chances of adverse reproductive outcomes. The risks of the most common adverse pregnancy outcomes likely to occur in women who postpone childbearing were tested in the general and IVF-treated populations.

7.1 Summary of the findings

Chapter 2 describes a matched case-control study based on national Danish health registry data. The study investigated whether women with a history of ovarian surgery, as a proxy for diminished ovarian reserve, had a higher risk of a trisomic pregnancy. Cases were women with a confirmed trisomic pregnancy occurring between January 1st 2000 and December 31st 2010 and controls were women with a live born child without a trisomy. In total, 1723 cases and 6850 age-matched controls were included in the study. A history of ovarian surgery was present in 2.7% (46/1723) of the cases versus 2.5% (172/6850) of the controls and was not associated with a higher risk for a subsequent trisomic pregnancy (OR 1.00, 95% CI 0.99-1.01). Subgroup analyses by indication of ovarian surgery and interval between ovarian surgery and pregnancy did not change the results.

Chapter 3 describes an age-matched case-control study investigating the risk of a trisomic pregnancy in relation to oocyte yield in IVF-treatment. The proxy for diminished ovarian reserve in this study was low oocyte yield (three or less oocytes retrieved after controlled ovarian stimulation). Cases were women with a trisomic pregnancy (trisomies 13, 18 or 21) resulting from fresh IVF treatment and confirmed by karyotyping. Controls were age-matched women with a live born child without a trisomy. Analyses were performed in 103 cases and 432 controls from Dutch and Danish national cohorts containing data obtained from IVF registries from 1983 to 2010. Low oocyte yield was observed in 6.5% (28/428) of the women, of whom 8.6% (7/81) were cases and 6.1% (21/347) were controls. Low oocyte yield was not associated with a higher risk of trisomic pregnancy (OR 1.2, 95% CI 0.8-1.9). Stratification for female age, adjustment for history of ovarian surgery and stimulation protocol used did not change the results.

Chapter 4 is a cohort study aimed to determine whether diminished ovarian reserve, reflected by repeated low oocyte yield in IVF-treatment, is associated with miscarriage. We included 933 Dutch IVF patients who had their first two IVF cycles
within one year and conceived after the second cycle. Patients were selected from a national Dutch IVF registry from 1983 to 1995. The proxy for diminished ovarian reserve in this study was the retrieval of three or less oocytes in both the first and second IVF cycle, i.e. a repeated cycles with low oocyte yield. Women achieving an ongoing pregnancy (viable intra-uterine pregnancy ≥ 16 weeks) were compared to those experiencing a miscarriage (pregnancy loss between 4-16 weeks). We analysed miscarriage risk in women with twice (4.6%), once (16.9%), and no (78.4%) cycle with low oocyte yield. Women with two cycles with low oocyte yield more often experienced miscarriage (37.2%) compared to women without low oocyte yield (18.7%) (crude OR 2.57, 95%CI 1.35-4.91; adjusted for female age OR 2.21, 95%CI 1.14-4.28). Women with one cycle with low oocyte yield did not have an increased risk of miscarriage (20.9%) compared to women without low oocyte yield, irrespective of whether low oocyte yield occurred in the first or second IVF cycle.

Chapter 5 is a cohort study including 7160 participants from the British ALSPAC study, a cohort in which women from the Avon County were included and followed-up. The proxy for diminished ovarian reserve in the study was in-utero cigarette smoke exposure of the participants. The adverse reproductive outcome was miscarriage, defined as a pregnancy loss after 20 weeks gestation or less. We used obstetric history data, obtained from medical records and questionnaires, regarding pregnancies occurring before inclusion of participants in the cohort in 1992. Participants who were exposed to cigarette smoke in-utero (n= 2049/7160, 28.6%) had a higher risk of ever having a miscarriage (33.7% versus 30.9%; adjusted OR 1.12, 95%CI: 1.00, 1.27) compared with participants who were not exposed. The risk of one previous miscarriage was not influenced by being in-utero exposed (OR 1.08, 95%CI: 0.95, 1.23). However, the risk of having two (OR 1.30, 95%CI: 1.02, 1.67) or three or more (OR 1.17, 95%CI: 0.80, 1.69) previous miscarriages was higher for participants who were exposed in-utero. There was an interaction between in-utero smoke exposure and three factors: packyears of the participants (i.e. the smoking behaviour of the participants themselves), passive smoke exposure from other household members, and age at first and last pregnancy of the participants. Participants exposed in-utero who were ever smokers themselves had an increased risk of miscarriage (OR 1.26, 95% CI: 1.11, 1.52) while participants exposed in-utero who were never smokers did not (OR 0.93, 95% CI: 0.78, 1.12).

Chapter 6 is also a cohort study within the ALSPAC cohort aimed to evaluate whether women exposed to cigarette smoke in-utero reach menopause earlier compared to non-exposed women. Women who were still active participants in the ALSPAC cohort in 2010, i.e. 18 years after enrolment period, were eligible for this study. There were 2852 participants eligible for analysis, of which 24.4%
(695/2852) were postmenopausal. Of all participants, 20.2% (577/2852) were exposed to cigarette smoke in-utero. In-utero smoke exposure was not associated with earlier menopause. Participants who were in-utero exposed but were not smokers themselves did not have higher hazards of menopause (adjusted HR 0.92, 95%CI 0.72-1.18), while participants who were ever smokers (current or previous) did, whether they were in-utero exposed (adjusted HR 1.41 95%CI 1.01-1.95) or not (adjusted HR 1.24 95% CI 1.00-1.53).

Figure 7.1: Summary of the findings in this thesis concerning an association between diminished ovarian reserve and adverse reproductive outcome

7.2 Discussion of the results of this thesis and studies available in the literature

In this thesis, ovarian surgery, low oocyte yield in IVF with controlled ovarian
stimulation and in-utero cigarette smoke exposure were taken as proxies for diminished ovarian reserve. Our results indicate that repeated low oocyte yield, defined as three or less oocytes retrieved in IVF treatment, and in-utero cigarette smoke exposure are associated with miscarriage (see figure 7.1). Moreover, smoking women who are exposed to cigarette smoke in-utero have a higher risk of earlier menopause.

### 7.2.1 Ovarian surgery as proxy for diminished ovarian reserve

Ovarian surgery was not associated with an increased risk of a trisomic pregnancy in our study, which contradicts two previous studies (OR for trisomic pregnancy 9.61, 95%CI 1.18-446.3) (Freeman et al., 2000) and (OR for trisomic pregnancy 3.3, 95% CI 1.0-10.5) (Haadsma et al., 2010b). These studies have smaller sample sizes and larger confidence intervals compared to the study in this thesis.

The effect of the reduction of ovarian reserve after ovarian surgery is demonstrated by studies showing an earlier menopause for women with unilateral oophorectomy (Cramer et al., 1995; Yasui et al., 2012; Bjelland et al., 2014) compared to women with both ovaries and studies showing a reduction in the results of ovarian reserve tests (ORTs) for women who had part of their ovarian tissue removed, for example, after excision of endometrioma (Almog et al., 2010).

However, systematic reviews evaluating the effect of surgery on ORTs are conflicting (Raffi et al., 2012; Somigliana et al., 2012; Muzii et al., 2014; Cagnacci et al., 2016). These may be explained by the fact that, first, the results of ORTs might be low already prior to surgery, due to the underlying pathology, such as endometriosis (Muzii and Miller, 2011; Lind et al., 2015). Second, the interval between surgery and ORTs assessments might have been different between studies and this interval seems to be important. ORTs seem to show lowest values immediately after surgery (kitajima et al., 2011), but exhibit partial recovery in longer follow-up assessments (Chang et al., 2010; Zhai et al., 2012; Sugita et al., 2013). It is hypothesized that this recovery can be caused by a compensatory mechanism of the remaining ovarian tissue. This may also be the explanation for the fact that women with previous oophorectomy have, on average, age at menopause only one year earlier, which is not proportional to the amount of ovarian tissue lost (Cramer et al., 1995; Yasui et al., 2012; Bjelland et al., 2014). In mice, after the removal of one ovary, the remaining ovary showed increased ovarian weight due to hypertrophy and increased formation of corpus luteum 19-21 days after surgery (Bhattacharya, 2013), but no changes in the number of primordial follicles after surgery (Aydin et al., 2010). In addition, although women with a single ovary have a lower response to ovarian stimulation (Lass et al., 1997b; Almog et al., 2010) and may need higher gonadotropin dosages (Lass et al., 1997b)
it was shown that the oocyte yield per ovary was higher in women with a previous oophorectomy compared to women with both ovaries in situ (Khan et al., 2014). These findings indicate that the loss of ovarian reserve after surgery is at least partly compensated by optimized use and function of the remaining ovarian tissue. One suggested mechanism is the fact that ovarian surgery may induce fragmentation of the cortex of the ovary. Ovarian fragmentation seems to waken dormant follicles via growth factors activated by the so called PTEN (phosphatase with tensin homology deleted in chromosome 10) inhibitor and PI3 kinase (phosphatidylinositol-3-kinase) stimulator (Hsueh et al., 2015). Ovarian fragmentation is being explored as a possible approach for infertility treatment (Kawamura et al., 2013; Suzuki et al., 2015). Other mechanisms could be the decrease of follicular atresia of FSH dependent follicles, possibly as a consequence of higher gonadotropin levels after oophorectomy and/or an increase in the recruitment or transition of the NGF (non-growing follicles) into the FSH dependent follicle pool. Ovarian surgery thus diminishes ovarian reserve via the direct mechanical removal of ovarian tissue, specifically when it involves the ovarian cortex, where the majority of the NGFs are located, but the effect of this reduction might mainly be temporary due to compensatory mechanisms. However, there is no strong evidence of lower quality of the oocytes as a result of ovarian surgery. In our study we could not confirm the limited pool hypothesis for the association between reduced oocyte quantity resulting from ovarian surgery and oocyte quality, using trisomic pregnancy as a proxy.

In support of our findings, embryo quality does not seem to differ (Harada et al., 2015) and live birth rates are similar in women with one ovary as compared to women with both ovaries (Lass et al., 1997a; Khan et al., 2014). Additionally, no difference was found in miscarriage rates between women who had unilateral ovariectomy (28.5%), and controls with cholecystectomy (30.8%) and appendectomy (28.2%) (Bellati et al., 2014). Therefore, we suggest that ovarian surgery has an effect on quantity, but not on quality of the oocytes.

**Conclusion:** Ovarian surgery, as a proxy for diminished ovarian reserve, has a negative effect on quantity of ovarian reserve, but this effect may be largely temporary. The adverse reproductive outcomes associated with ovarian surgery are likely to be restricted to outcomes associated with the quantity of follicle pool, such as earlier menopause, and not quality, such as trisomic pregnancy or miscarriage.

**Clinical implications:** Women with a history of ovarian surgery prior to pregnancy may not be regarded to be at a higher risk for a trisomic pregnancy or miscarriage. In case these women need IVF treatment, higher dosages of ovarian stimulation...
may be needed, and a lower oocyte yield might be anticipated. Their embryos are not likely to have lower quality, and the chances of live birth are similar to women with two ovaries. However, these women need to be aware that their reproductive lifespan might be in average, one year shorter. That needs to be taken into consideration when planning when to start a family.

7.2.2 Low oocyte yield as proxy for diminished ovarian reserve

Low oocyte yield in IVF treatment was not associated with a higher risk of having a trisomic pregnancy in our study. This finding differs from a previous study from our research group (Haadsma et al., 2010b), in which the odds for a trisomic pregnancy for women with three or less oocytes retrieved was around three times higher compared to women with normal response (OR for trisomic pregnancy for women with three or less oocytes retrieved 2.72, 95% CI 0.69-10.69) (Haadsma et al., 2010b). This difference in results is attributed to differences in study population and statistical analyses. The data from the Haadsma et al. study were re-analysed with the statistical techniques used in the study in this thesis, and the differences were no longer present. We concluded that the results of the previous study are likely to show an overestimation of the effect.

Within the same patient, the number of recruited follicles differs per cycle (Elter et al., 2005; van Disseldorp et al., 2010). A low oocyte yield after controlled ovarian stimulation might be the result of this inter-cycle variability or may result from underdosage of gonadotropins. In this situation, there is no diminished ovarian reserve: a sufficient total number of follicles is available, but only a few are recruited in that particular cycle. A different situation is when the number of follicles is actually diminished, in which no matter the stimulation dosage, type or cycle number, the response will always be low. A second cycle with low oocyte yield occurs in 36% of women who had a previous cycle with low oocyte yield (Klinkert et al., 2004). Therefore, the majority of women with a low oocyte yield after controlled ovarian stimulation might not have a diminished ovarian reserve.

Two consecutive cycles of low oocyte yield (or more) is a condition in which the patient is called a “poor responder”. In addition to two cycles of low oocyte yield, the Bologna criteria define a “poor responder” as a patient with at least two of the following criteria: (a) age > 40 years, (b) previous cycle with a low oocyte yield or (c) abnormal ovarian ORTs (AMH < 0.5-1.1 ng/ml or AFC < 5-7 follicles) (Ferraretti et al., 2011). Therefore, being a “poor responder” is likely to be a better proxy for diminished ovarian reserve than one cycle of low oocyte yield alone. In this thesis, women with two consecutive cycles of low oocyte yield were at a higher
risk for miscarriage, one of the adverse outcomes presumably resulting from low oocyte quality. A single response of three or less oocytes after controlled ovarian stimulation is not necessarily associated with poor outcomes such as lower embryo quality or lower live birth rates (Arce et al., 2014; Mustafa et al., 2017). But a low oocyte yield, in women fulfilling the Bologna criteria for “poor responders” is more indicative of poor prognosis (Busnelli et al., 2015; Marca et al., 2015). Live birth rates for “poor responders” are expected to be as low as 6-10% (Polyzos et al., 2014; Busnelli et al., 2015; Marca et al., 2015).

Being a poor responder might well be a condition in which the quantity and quality of the ovarian reserve are associated. The literature indicates that one cycle of low oocyte yield is associated with earlier menopause (de Boer et al., 2003; Lawson et al., 2003; Szmidt et al., 2016) and there are studies indicating an association between one cycle of low oocyte yield and adverse reproductive outcomes (Haadsma et al., 2010a; Sunkara et al., 2014). This effect might be related to the “poor responders” included within these groups of women with low oocyte yield. This might be particularly true when the cut-off for low oocyte yield is set at three oocytes or less. Once the cut-off is elevated, e.g. to five oocytes or less, the proportion of “poor responders” within the group of women with low oocyte yield decreases, and the effect of low oocyte yield on adverse reproductive outcome is no longer observed (Kumbak et al., 2009; Setti et al., 2011).

**Conclusion:** The effect of low oocyte yield as a proxy for diminished ovarian reserve is not straightforward because of the “false positives” included in that group, i.e., women with a normal ovarian reserve experiencing a random low oocyte yield. While there was no effect of one cycle of low oocyte yield on trisomic pregnancy risk, once the proxy was a “poor responder”, defined as women with two consecutive cycles of low oocyte yield, we observed a higher risk for miscarriage. Women who are “poor responders” are the ones at higher risk for adverse reproductive outcomes. Thus, being a “poor responder” is a condition in which the quantity and quality of the oocytes are associated.

**Clinical implications:** A low oocyte yield in a first cycle should be interpreted with caution. Low oocyte yield is not necessarily associated with a diminished ovarian reserve. After a first cycle with low oocyte yield, patients should be evaluated using the Bologna criteria. If they fulfil the criteria, they should be counselled of a poor prognosis for a live birth. Women achieving a live birth after being diagnosed as a “poor responder”, she should be counselled in case there is a desire for another pregnancy, that the interval between pregnancies should be the shortest appropriate interval in order to minimize the effects of further decline of ovarian reserve, quantitatively and qualitatively with time.
7.2.3 In-utero cigarette smoke exposure as proxy for diminished ovarian reserve

In-utero cigarette smoke exposure, as a proxy for diminished ovarian reserve, was associated with a higher risk of having a miscarriage in our study. This is in accordance with the results from Cupul-Uicab and colleagues (Cupul-Uicab et al., 2011), the only other study available on this subject. This is suggestive that cigarette smoke exposure in-utero has an effect on the quality of the foetal oocytes. With respect to the quantity of ovarian follicles, animal studies showed a decreased number of primordial and primary follicles and ovarian volume in mice exposed to cigarette smoke in-utero (Sobinoff et al., 2013; Camlin et al., 2016). The only human study with foetal ovaries showed a decreased number of oogonias in foetuses who were in-utero exposed to cigarette smoke (Lutterodt et al., 2009). So indeed, in-utero cigarette smoke exposure leads to a lower ovarian reserve early in life.

With respect to quality of ovarian follicles, meiotic errors due to alterations in spindle formation were observed in mice exposed to cigarette smoke in-utero (Camlin et al., 2017). These alterations are associated with aneuploidies in oocytes (Eichenlaub-Ritter, 2012) leading to higher risk of trisomic pregnancy and miscarriage. Summarizing, these studies show that cigarette smoke exposure in-utero has an effect on ovarian reserve both quantitatively and qualitatively.

However, in-utero cigarette smoke exposure, as a proxy for diminished ovarian reserve, was not associated with a higher risk of having an earlier menopause in our study. There are three previous studies on this same issue but with conflicting results (Strohsnitter et al., 2008; Steiner et al., 2010; Tawfik et al., 2015). The strongest evidence, from the study of Steiner and colleagues, with a large sample size (n=22165) and appropriate statistical method to analyse the research question (Steiner et al., 2010), suggests that there is no effect of in-utero smoke exposure on age at menopause. Therefore, in-utero cigarette smoke exposure may not be a proper proxy for diminished ovarian reserve and seems to have an effect on oocyte quality but not on quantity.

This is an opposite conclusion from the effect of ovarian surgery, in which there seems to be no effect on oocyte quality, but which does affect oocyte quantity, though less than expected, probably mediated by compensatory mechanisms. These compensatory mechanisms, however, might be a general response to ovarian insult, and it could also explain the lack of difference in age at menopause for women who were exposed in-utero and those who were not. Putatively, the less ovarian tissue a woman has, the more efficient the compensatory mechanism to
spare unnecessary follicle loss might be. The pathway however, might be different.

In this thesis cigarette smoke exposure in-utero alone had no effect on age at menopause, but there was an interaction cigarette smoking later in life: women who were smokers themselves and had been exposed in-utero did have earlier menopause, which was also observed in a previous study (Tawfik et al., 2015). The same interaction was observed when miscarriage was the adverse outcome, i.e. miscarriage risk was increased for women who were smokers and had been exposed to cigarette smoke in-utero. Possibly, the compensating mechanisms of the ovary fail when being hit with the toxic effects of smoking twice. First in-utero, leaving the ovaries with less and arguably partly damaged oocytes to start out with, and second when the woman starts smoking herself. But exactly what occurs to the dynamics of ovarian, follicle recruitment and follicle loss once there is a disturbance early in life to explain this interaction is not clear. The increased miscarriage risk that smoking women face is not likely to be caused by an increase in aneuploidies; many studies indicate that trisomic pregnancies are not associated maternal smoking (Rudnicka et al., 2002). There is, however, a gap in the literature with respect to cigarette smoke exposure in-utero and a higher chance of having a trisomic pregnancy later in life. It is perceivable that the toxic effects of smoking on the developing human ovary in-utero may differ from the effects of smoking of the woman herself when her ovaries are full-grown. A higher chance of trisomic pregnancies was indeed established in mice after in-utero smoke exposure. However, a note of caution is necessary for the direct translation of the results to humans since the foetal ovarian development in humans differs from that in mice. Unlike in humans, in mice, the development of primordial follicles proceeds after birth (Chadio and Kotsampasi, 2014)

**Conclusion**: Cigarette smoke exposure in-utero, as a proxy for diminished ovarian reserve, seems to be associated with diminished quality of the oocytes, leading to an increased miscarriage risk later in life. However, we don’t know whether cigarette smoke exposure in-utero is actually an optimal proxy for a decrease in quantity, as menopausal age is not affected. Thus, the relation between quantity and quality cannot be confirmed in the studies in this thesis.

**Clinical implications**: Smoking during pregnancy affects the reproductive capacity in two generations. The reproductive lifespan of the daughters might not be affected, but there is an increased risk of miscarriage for in-utero exposed daughters later in life. This is another strong argument, on top of many others, not to smoke, especially in the preconception period.
7.3 General conclusion- diminished ovarian reserve and adverse reproductive outcomes

Diminished ovarian reserve is a result of various mechanisms leading to a decrease in the quantity of the oocytes. First of all, diminished ovarian reserve is caused by the physiological effects of ageing. This results in both a decline in quantity and quality of the oocytes; two processes that seem highly correlated. The Bologna criteria for “poor responders”, also used in this thesis, show parallels with diminished ovarian reserve as caused by age, i.e., both quantity and quality of oocytes are affected and related. Differently, and with opposite effects in the outcomes described in this thesis and related literature, ovarian surgery showed an effect on quantity of oocytes only, without effect on oocyte quality and cigarette smoke exposure in-utero showed an effect on quality of the remaining oocytes only, without effect on menopausal age.

There is no consensus in the literature on the definition of diminished ovarian reserve, neither on which of its features are associated with adverse reproductive outcomes (Cohen et al., 2015). This thesis suggests that the underlying mechanism leading to a diminished ovarian reserve will determine whether there is a clinical detectable association between oocyte quantity and quality, reflected by adverse reproductive outcomes. Moreover, little is known about the presumed compensatory mechanisms within the ovary that might add to the effects.

7.4 Reflections on the use of epidemiological studies

This thesis explored a possible association between diminished ovarian reserve and adverse reproductive outcomes via epidemiological retrospective observational and case-control studies. Recall bias and misclassification bias are limitations for these types of studies. When possible, we tried to reduce the possibility of these kinds of bias by using and analysing repeated questions in questionnaires and comparing the information from medical files. We dealt with missing data properly in order to diminished the chances of biased results. We performed sensitivity analyses in the various studies to evaluate the effect of groups excluded from the analysis or specific groups within the data.

The major confounder in all our studies is age and was addressed properly by analysis with adjustment for age, stratification and matching by age. However, the best statistical methods will never be able to overcome the limitations of using proxies. Ovarian reserve cannot be studied in vivo. We used proxies for the association between oocyte quantity and oocyte quality. Perfect biological markers for ovarian reserve and oocyte quality do not exist. The limitations of our proxies for low oocyte quantity are discussed above. In addition, not all miscarriages and
trisomic pregnancies are the result of poor oocyte quality (Hunt and Hassold, 2010; Nagaoka et al., 2012; Jia et al., 2015). However, when oocyte quality is affected, these adverse reproductive events are most prevalent clinically relevant and detectable outcomes.

7.5 Future perspectives

We used epidemiological studies to assess the underlying relations and conclude that, putatively, the underlying mechanisms leading to a diminished ovarian reserve are associated with various adverse outcomes. We observed a gap in the literature concerning cigarette smoke exposure in-utero and adverse reproductive outcomes, such as trisomic pregnancies, congenital anomalies, but also fertility of the daughters and sons. With big data and birth records becoming more common, such studies become feasible and they should be less complicated to design. Another question regarding cigarette smoke exposure in-utero is the possibility of an effect on the grandchildren of the smoking mothers, since the grandchildren are already present as gametes in-utero, including their reproductive potential. Future studies should consider the mechanisms underlying diminished ovarian reserve in order to hypothesize for an existing association with quality. Basic research is necessary to explore the various underlying biological mechanisms leading to the decline in the ovarian follicle pool and decrease in oocyte quality. In general, relatively little is known about the dynamics of follicle atresia and recruitment. Concerning oocyte quality, studies on important regulatory role of mitochondria in oocytes, but also the surrounding granulosa cells, show that with age and exposure to specific diet and environmental toxins, their regulatory role is jeopardized leading to cell cycle abnormalities, meiosis defects and aneuploidy (Babayev and Seli, 2015). Moreover, the presumed compensatory mechanisms leading to less devastating effects of ovarian surgery on ovarian reserve can be further explored in animal models. Knowing which genes and products are either up- or downregulated in the remaining ovary after unilateral oophorectomy may provide insight in how a reduced number of follicles is used optimally. These intra ovarian compensatory mechanisms may have a therapeutic potential to aid infertile patients and women who are at risk for shorter reproductive lifespan. The process of ovarian ageing is as yet irreversible but maybe with better fundamental knowledge on its decline, we can perform some damage control.
7.6 Final reflections

The Declaration on Population of the United Nations declares that it is the choice of each individual to determine its family size (Ayala and Caradon, 1968). Free choice is illusory unless alternative possibilities are known. Unfortunately, most women are unaware that delaying childbearing increases the chances of involuntary childlessness (Präg et al., 2017).

Sexual education programs are mandatory in many countries in Europe for already many decades. It has been introduced as early as 1956 in Sweden and in the 1970’s for most other countries (Bott, 2013). The content of these programs covers unwanted pregnancy, sexually transmitted diseases, abortion and contraception. Biological aspects such as sperm production, ovulation and fertilization are also explained. However, changes in reproductive fecundity with age are not discussed (Nargund, 2015). The studies included in this thesis indicate that different conditions of diminished ovarian reserve have an impact either on adverse reproductive outcomes or reproductive lifespan. Moreover, it should be taught that IVF treatments cannot overcome conditions of diminished ovarian reserve. Additional to the traditional orientations received in schools regarding prevention of pregnancy, young women should know the effect of age and other conditions of diminished ovarian reserve on their fecundity. Once aware of the limitations of ovarian physiology, women are better equipped to decide when to start a family.

However, it is cruel to disregard the social context and blame the individual choice for unwanted childlessness. In most professions, starting a family functions as a hold-back-penalty for women. In the academic field for example, compared to a male PhD, a female PhD is 38% less likely to enter a tenure track when becoming a parent prior to 5 years post-PhD (Mason and Goulden, 2002). On the other hand, women that do have children earlier in life are highly likely to be less educated and fall into the “fertility poverty trap”, which is the observed association between early motherhood and child poverty (Dixon and Margo, 2000).

Societal support and change of concept can be of great help to diminish female struggle in this area. For example, many countries have compulsory military service for men. In such countries, the societies are organized to allow men to devote one or more years to learn basic military skills, and once this period is completed, men return to their normal lives, sometimes with some advantages, like a basic salary, or increase in pension. Although this compulsory character of military services is changing to a voluntary character, this is a clear example of socially built acceptance governmental policy. Similar acceptance has to be built within societies on with regard to maternity and paternity leave, child care and return to work support. This may help couples to fulfil their wish to have the number of children they desire, but may also add in general to improve birth rates.
Some countries are already taking action. Policies adopted by some governments to increase birth rates include family allowance, paternity leave, baby bonus, child tax allowance, subsidized child care among others (UN, 2011). These policies increase labour supply for women in all educational levels and increase the probability of having a child in 5-17% depending on country and policy (Boca, 2015). In Austria, for example, the chance of having a second child increased 15% with an additional year of parental leave benefits (Haan and Wrohlich, 2009).

However, policy makers must be cautious not to jump from birth limiting measures to an all-too-active pro-natal policy. The issue now is lack of awareness of female fecundity with increasing age and lack of social-economic gender equality, which should be addressed first. If and when couples try to start and have a family must remain a personal choice, made consciously and well-informed.