



Review

# Ovarian Telomerase and Female Fertility

Simon Toupance <sup>1,†</sup> , Anne-Julie Fattet <sup>2,†</sup>, Simon N. Thornton <sup>1</sup>, Athanase Benetos <sup>1,3</sup>, Jean-Louis Guéant <sup>4</sup> and Isabelle Koscinski <sup>2,4,\*</sup>

<sup>1</sup> DCAC, Université de Lorraine, Inserm, 54000 Nancy, France; s.toupance@chru-nancy.fr (S.T.); simon.thornton@univ-lorraine.fr (S.N.T.); a.benetos@chru-nancy.fr (A.B.)

<sup>2</sup> Laboratory of Biology of Reproduction, University Hospital of Nancy, 54000 Nancy, France; annejulie1208@hotmail.fr

<sup>3</sup> Université de Lorraine, CHRU Nancy, Pôle “Maladies du Vieillissement, Gériologie et Soins Palliatifs”, 54000 Nancy, France

<sup>4</sup> Inserm U1256, NGERE, Université de Lorraine, 54000 Nancy, France; jean-louis.gueant@univ-lorraine.fr

\* Correspondence: koscinski.isa@gmail.com; Tel.: +33-624-263-768

† These authors have contributed equally to this work.

**Abstract:** Women’s fertility is characterized both quantitatively and qualitatively mainly by the pool of ovarian follicles. Monthly, gonadotropins cause an intense multiplication of granulosa cells surrounding the oocyte. This step of follicular development requires a high proliferation ability for these cells. Telomere length plays a crucial role in the mitotic index of human cells. Hence, disrupting telomere homeostasis could directly affect women’s fertility. Strongly expressed in ovaries, telomerase is the most effective factor to limit telomeric attrition and preserve ovarian reserve. Considering these facts, two situations of infertility could be correlated with the length of telomeres and ovarian telomerase activity: PolyCystic Ovary Syndrome (PCOS), which is associated with a high density of small antral follicles, and Premature Ovarian Failure (POF), which is associated with a premature decrease in ovarian reserve. Several authors have studied this topic, expecting to find long telomeres and strong telomerase activity in PCOS and short telomeres and low telomerase activity in POF patients. Although the results of these studies are contradictory, telomere length and the ovarian telomerase impact in women’s fertility disorders appear obvious. In this context, our research perspectives aimed to explore the stimulation of ovarian telomerase to limit the decrease in the follicular pool while avoiding an increase in cancer risk.

**Keywords:** telomerase; hTERT; female fertility



**Citation:** Toupance, S.; Fattet, A.-J.; Thornton, S.N.; Benetos, A.; Guéant, J.-L.; Koscinski, I. Ovarian Telomerase and Female Fertility. *Biomedicines* **2021**, *9*, 842. <https://doi.org/10.3390/biomedicines9070842>

Academic Editor: Yegor E. Yegorov

Received: 5 June 2021

Accepted: 14 July 2021

Published: 20 July 2021

**Publisher’s Note:** MDPI stays neutral with regard to jurisdictional claims in published maps and institutional affiliations.



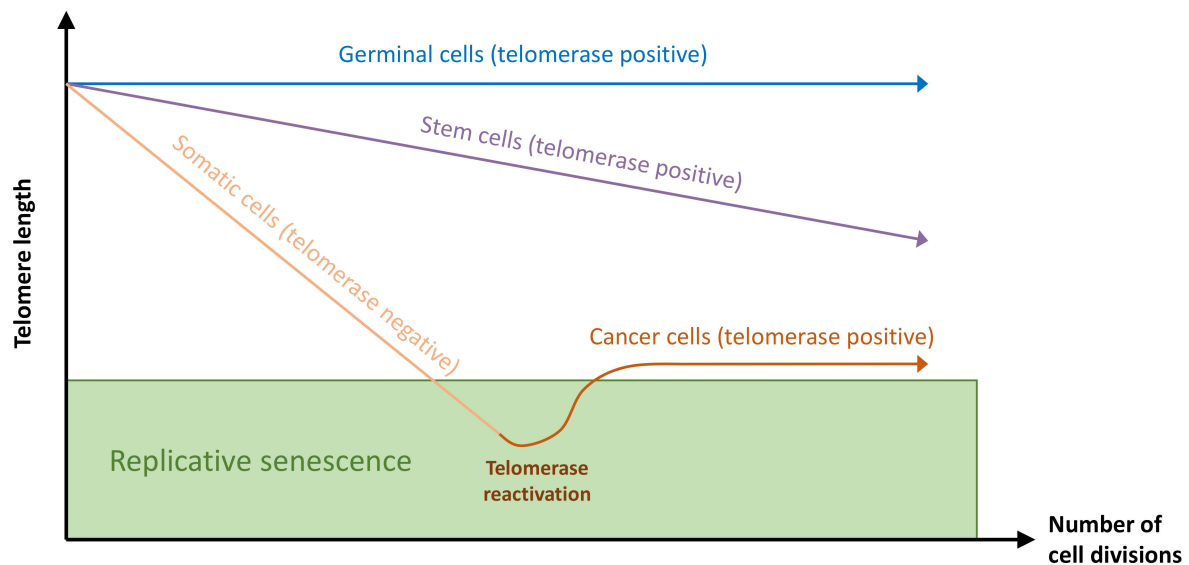
**Copyright:** © 2021 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (<https://creativecommons.org/licenses/by/4.0/>).

## 1. Introduction

Female fertility is strongly linked to the number of ovarian follicles and their quality. The pool of ovarian primordial follicles is established during prenatal life and results from an intensive proliferation of ovogonia and somatic follicular cells that surround them. Follicular atresia begins even before birth and ends with menopause. The quality of the follicles is linked to their capacity to ovulate, to produce a fertilizable oocyte that leads to an evolutionary embryo, and finally to a healthy child. Folliculogenesis implies an intense proliferation of granulosa cells during follicular maturation. Overall, the quantity and quality of follicles depend greatly on the capacity of follicular cells to proliferate; any disruption of this mechanism will result in a shortened reproductive life span and ovulation disturbance. Telomeres, the nucleoprotein structures that cap chromosome extremities, play a crucial role in the proliferative potential of cells. Telomere length shortens with each cell division and triggers cell senescence when a certain threshold is reached. Therefore, telomere attrition is considered a primary hallmark of aging [1]. This review attempts to highlight the relation between telomere length/telomerase activity in ovarian cells physiologically and in two situations of infertility, PCOS and POF.



with SP1 [32,33] and, through this way, silence the expression of some genes, especially tumor suppressors.



**Figure 2.** Telomere length dynamics in germinal, stem, somatic, and cancer cells according to the presence of telomerase activity. In germinal and stem cells, active telomerase can offset telomere attrition due to cell divisions. In somatic cells, telomerase is inactive, and telomere length is shortened until it reaches a threshold that triggers replicative senescence. Cancer cells can bypass cell senescence by the reactivation of telomerase.

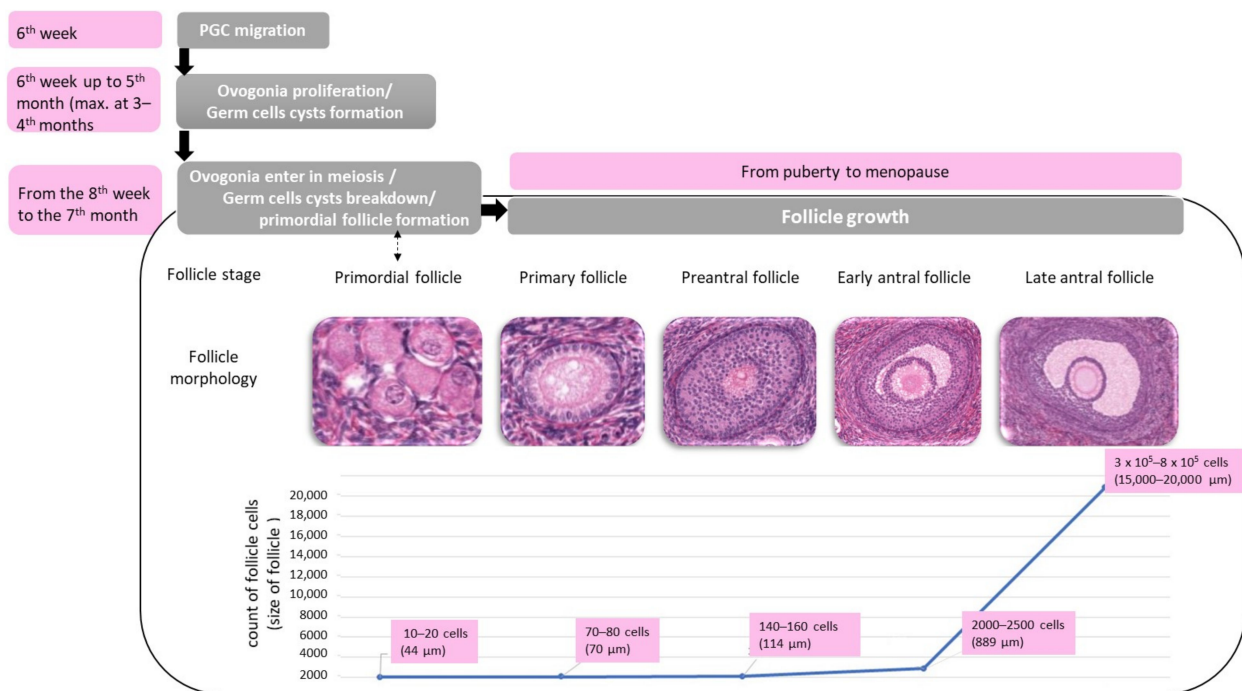
Under oxidative stress conditions, TERT can move out from the nucleus and localize to mitochondria [34,35]. In mitochondria, TERT can bind to mitochondrial DNA (mtDNA) and display a protective effect against reactive oxygen species, protecting cells against oxidative stress-induced damage [35–37]. TERT can also sensitize cells to oxidative stress by increasing levels of mtDNA damage, which may lead to apoptotic cell death [38]. This discrepancy in the mitochondrial role of TERT on oxidative stress can be explained by different levels of TERT expression. Mitochondrial TERT also exerts control on mitophagy by decreasing the processing of PINK1 (phosphatase and tensin homolog-induced putative kinase 1) [39]. TERC can also be imported into mitochondria [40], where it is converted to TERC-53 and exported back to the cytoplasm. TERC-53 acts as a signaling molecule involved in cell senescence [41].

### 3. Human Folliculogenesis, the Importance of Proliferative Capacity

Ovaries contain female gametes in special histologic structures named follicles, where the gamete (oocyte) is surrounded by somatic cells (follicular cells). Folliculogenesis comprises several steps summarized here and illustrated by Figure 3, with a special focus on granulosa cell proliferation.

#### 3.1. Establishment of a Pool of Primordial Follicles at the Fetal Time

After their migration into the genital ridges, primordial germ cells undergo many mitoses from the 6th week of embryo development, with a maximum mitosis rate in the 3rd and 4th month, leading to the formation of primordial germ cell cysts. From the end of the 2nd month, variable numbers of ovogonia enter meiosis and block at the diplotene stage of the first meiosis division [42]. At the same time, these germ cell cysts break apart into oocytes that become surrounded by somatic pre-granulosa cells to form primordial follicles whose number is maximal at the 5th month of pregnancy in humans [42]. All these steps are enabled by the high proliferation capacity of ovogonia and follicular cells [43] and may be limited by short telomeres, restricting the size of the follicular pool [44].



**Figure 3.** Human folliculogenesis and granulosa cells proliferation. The figure describes the different stages of follicle growth and the corresponding granulosa cell count and follicle size. Granulosa cells need to display a high proliferative potential to accommodate the tremendous proliferation needed during folliculogenesis.

### 3.2. Follicle Growth

From puberty until menopause, follicular development starts when follicles leave the pool of resting primordial follicles to enter the growth phase. From there, the early growing follicle will become dominant, undergoing a developmental process, including a dramatic course of cellular proliferation and differentiation [45]. This occurs similarly in all mammals: for example, bovine follicular cells of the primordial follicle divide 21 times until the stage of an antral follicle [46].

The next step of follicle maturation has also been well explored in bovine and implies a differentiation of granulosa cells into two different pools: granulosa cells building the wall of the antrum and granulosa cells forming the cumulus oophorus around the oocyte. In the antral wall, the proliferation rate is greater in the middle and antral layers than in the basal layer, where it seems similar, as observed in the cumulus oophorus [47].

This development of primordial into secondary follicles and subsequent maturation of the dominant follicle implies a very high proliferation capacity for granulosa cells [48]. The mitotic index of the granulosa cells continues to increase with follicular diameter before reaching a peak in the small antral follicles [45,49]; the granulosa cell number reaches to the hundreds of thousands before ovulation [50] (Figure 3).

### 3.3. Telomerase and Follicle Growth

In this context, the telomere length of ovarian cells has crucial importance [48]. As the most efficient factor limiting telomere attrition, the telomerase presents, unsurprisingly, rather high activity in the ovaries, especially in granulosa cells, which may enable their numerous replications. This high telomerase activity was highlighted many years ago in cows [47] and recalls other multipotent stem cell characteristics of granulosa cells [51,52]; that is, they divide without the need of anchorage [53] and can be differentiated into varied cell types [54].

The activity pattern of the telomerase has been studied through the different types of follicles in ovaries and with aging, especially in large mammals (mainly in bovines and pigs) [55,56].

Heterogeneity among all follicles has been found; the highest levels of telomerase activity are found in the smaller, rapidly growing preantral follicles [49,55], followed by a decrease in activity with the maturation of the follicle [48]. This suggests that the high proliferative activity of granulosa cells could be partially linked to telomerase activity [57].

Interestingly, the study of telomerase in bovine antral follicles highlighted heterogeneity within the granulosa, with a pattern similar to the proliferation index. More importantly, the expression of the catalytic subunit of TERT appears conserved in the middle and next to the antrum layers (layers with a high proliferation rate), whereas it seems to be down-regulated in basal layers and in the cumulus oophorus, where only residual telomerase RNA would persist [47,58], explaining a moderate proliferation rate during the last stage of follicle maturation. In pigs, comparable proliferation and TERT activity have been observed in granulosa cells of the wall of the antrum (expression of TERT only in middle and antral layers, and not in basal layers); however, TERT has also been detected in cumulus cells at a higher level than in bovine [56,59].

After ovulation, residual granulosa cells undergo differentiation to large luteal cells and enter apoptosis. Not surprisingly, they present a low telomerase activity. This evolution of granulosa cells, which have achieved their differentiation and are morphologically distinct from proliferating granulosa cells in early-stage follicles, can explain the decrease in telomerase activity, as reported in some studies [60].

### 3.4. Regulation of Telomerase Activity in Ovaries

#### 3.4.1. Effect of Age

Interestingly, aging significantly impacts ovarian telomerase activity in bovine [55], leading to a decrease in granulosa cells' proliferative activity. In mice, ovarian TERT expression was found to decrease with aging [61]. In humans, ovarian TERT also decreases with aging and is associated with telomere attrition [62].

#### 3.4.2. Estradiol and Telomerase Activity

The relation between sex hormone and telomere length has been considered for some time, based on observations of longer telomeres in women than in men [63]. The sex-specific discrepancy of the dynamics of telomere length erosion throughout life [64] was first hypothesized, suggesting a protective impact of female sex hormones [65]. However, longitudinal studies are controversial, showing either higher or lower rates of telomere length (TL) attrition in women compared to same-aged men [66,67]. Recently, it was even suggested that the sex-related gap in TL could be established at birth since gender differences in TL are found in newborns [68].

However, these longitudinal data originate from leukocyte telomere length (LTL) analysis, and tissue-specific differences in TL dynamics can exist. During folliculogenesis, in ovarian aging, as well as during neoplastic development, the pattern of induced telomerase activity is dependent on cell type, time, and location [56].

A direct impact of estrogens has been suggested since the region of the *TERT* promoter presents an estrogen response element [67]. An indirect impact via the activation of *MYC*, which is a direct target of ESR, has been suggested by studies in granulosa of rats and mice [69,70]. Activation of the proto-oncogene *MYC* seems especially involved in ovarian carcinogenesis [57], like *BCL2* and *TERT*.

In addition, high levels of estrogens can act indirectly on TL through a reduction in reactive oxygen species [71]. Furthermore, epidermal growth factor (EGF), which stimulates telomerase activity, has been found in abundance in small follicles [59], suggesting its role in the regulation of the telomerase activity and telomere maintenance, particularly in small, rapidly growing follicles.

The estradiol impact on telomerase and telomere length dynamics has been explored by animal experiments: in rats, the administration of estrogens prevents the inhibition of telomerase, suggesting that telomerase withdrawal plays an integral role in granulosa cell apoptosis and follicular atresia induced by estrogen withdrawal [72]. On the contrary,



estrogen deficiency in mice results in an inhibition of telomerase within ovaries in a tissue-specific manner. This leads to the shortening of telomeres and compromised proliferation of the follicular granulosa cells. These effects are offset by estrogen replacement therapy [73]. Interestingly, in return, telomerase could exert control on estrogen expression through the non-canonical pathway. A recent study showed that *in vitro* overexpression of TERT in granulosa cells enhanced the expression of steroidogenesis genes and increased the level of estrogen in pre-ovulatory luteinized granulosa cells [74].

Considering the importance of estrogen in the regulation of telomere length, especially through the regulation of telomerase activity, two physio-pathological situations can be studied from a telomerase point of view: the syndrome of polycystic ovaries (PCOS) relative to a very high ovarian follicular density, and high estrogen levels and premature ovarian failure (POF) relative to a very poor ovarian follicular density and poor estrogen levels. Based on the elements exposed above, long telomeres with high telomerase activity in PCOS patients and short telomeres with decreased telomerase activity in POF patients would be expected to be found.

#### 4. Polycystic Ovary Syndrome (PCOS)

The syndrome of polycystic ovaries is the most common endocrine disorder causing female infertility. Among women of reproductive age, its prevalence is reported to be 6–8% in developing countries. PCOS is a multifactorial disorder, and symptoms include hyperandrogenemia, oligo-anovulation, and a polycystic morphology of the ovaries. In addition, oocyte quality is frequently impaired, and, finally, PCOS constitutes real ovulatory infertility. Furthermore, some metabolic disorders such as metabolic syndrome, type 2 diabetes, and cardiovascular disease are regularly associated with PCOS [75] due to a high incidence of insulin resistance (IR), which participates in the pathophysiology of this syndrome. The link between IR and sex hormones has been studied for over 20 years. IR is promoted more by high estrogen levels than high androgen levels, and hyperinsulinism seems to promote hyperandrogenism. Indeed, insulin can stimulate ovarian sex hormone secretion directly by linking to the ovarian insulin receptor and IGF-I receptor, and indirectly mainly by reducing the hepatic production of IGFBP-1 (the major circulating IGF-1 protein) [76,77]. PCOS is also associated with a proinflammatory state [78] since oxidative stress is not compensated by antioxidative factors [79]. The main symptoms and long-term conditions of PCOS are provided in Table 1.

**Table 1.** Diagnostic criteria according to AE-PCOS Society (2006) and long-term consequences [77].

	Clinic and Biologic Symptoms	Long-Term Clinical Consequences
Diagnostic criteria according to Androgen Excess-PCOS Society (2006)	Biochemical and clinical hyperandrogenism (hirsutism and acne)	Hirsutism
	Ovarian dysfunction (oligo-amenorrhea)	Infertility
	Polycystic ovary morphology	
	Insulin resistance/increased incidence of type 2 diabetes	Metabolic syndrome
Increased incidence of cardiovascular disease		

PCOS pathophysiology is quite complex, and precise etiology remains to be elucidated. It results from environmental and genetic factors with a high degree of heritability. No single factor can fully explain the spectrum of abnormalities in PCOS; some studies reported that it might be related to longer telomere length in granulosa cells [75]. It has also been suggested that the persistent oxidative stress observed in PCOS may influence telomere homeostasis. Therefore, high inflammatory factors, especially reactive oxygen species, may increase telomeric attrition [80] and impair telomerase activity [81]. Telomere

shortening may be another mechanism associated with the pathogenesis of PCOS and its comorbidities. In response to gonadotrophin treatment, PCOS patients frequently develop many follicles, but these follicles often contain oocytes of small size and with a low rate of maturation (metaphase II). Moreover, metaphase II oocytes from PCOS patients frequently present an impaired competence. Since the interaction between oocyte and surrounding granulosa cells is crucial for the maturation and acquisition of oocyte competence, oocyte abnormalities could result from an impaired interaction between oocyte and granulosa cells and/or an insufficient proliferation of granulosa cells. Insofar as telomere length and telomerase activity play a major role in folliculogenesis, some authors propose that telomere disruption and limited activity of telomerase might be involved in the PCOS syndrome [82].

Studies on the relation between TL and PCOS are contradictory. Many have focused on the peripheral blood leukocytes of PCOS patients and reported shorter LTL [78,83]. However, tissue-specific TL dynamics can exist, especially in ovaries that express telomerase activity.

Recently, Pedroso et al. [82] measured telomere length and telomerase activity in the ovarian cells of women with PCOS in comparison with women with regular menstrual cycles (control), who underwent in vitro fertilization with intracytoplasmic sperm injection (IVF/ICSI). They found no difference in the telomere length of cumulus cells from PCOS and non-PCOS women. However, telomerase activity in cumulus cells was higher in the PCOS group. In the immature oocytes (germinative vesicle and metaphase I), telomere length and telomerase activity did not differ between the two groups. Interestingly, telomere length in leukocytes was shorter in PCOS patients than in controls. The authors proposed that telomere length in cumulus cells from PCOS patients might be maintained thanks to a higher telomerase activity [82]. Moreover, the authors reported a positive correlation between androgen levels and telomerase activity in cumulus cells and concluded that androgens stimulated telomerase [82]. The mechanism by which androgens stimulate telomerase activity could involve aromatization and be linked to  $E\alpha$ , as proposed in hematopoietic cells [84]. The role of the high production of estrogen, characteristic of PCOS, is also very probable. Thus, androgens and estrogens contribute to telomere maintenance by increasing telomerase expression and activity.

Using a similar study design, Li et al. [85] found shorter granulosa cell TL but similar telomerase activity levels in PCOS patients. These observations suggest that the impact of other factors such as insulin resistance or oxidative stress on TL is important. However, the authors showed that patients with lower TA and shorter TL exhibited a longer duration of infertility and earlier onset of infertility symptoms [85]. This suggests that a reduction in TA in granulosa cells might be associated with ovarian function decline [57].

In contrast, Wei et al. [75] demonstrated significantly longer GTL in PCOS patients. This finding reinforced the possible impact of PCOS high hormone levels on telomere homeostasis [75]. Furthermore, as described above, higher telomerase activity is observed in the smallest preantral follicles, which are six times more abundant in PCOS ovaries than in other patients [47,72,86].

To conclude, the results of the different studies are controversial, and they are summarized in Table 2. Considering the variability of phenotypes associated with PCOS, many factors are implicated in the pathophysiology of this syndrome. Although these studies demonstrate evidence of telomeric disruption in PCOS, the hyperandrogenism present in these women may compensate for the deleterious effect of the inflammatory state and metabolic disorders related to PCOS. To identify and clarify the implication of telomeric homeostasis in PCOS, further studies are required, with flawless methodology conducted on larger populations and using the most suitable method of TL measure.

**Table 2.** Studies measuring telomere length/telomerase activity in granulosa cells in PCOS.

Authors	Variable Measured/Method	Population No.	Population Age (Years)	Results
Li et al., 2017 [85]	GTL: PCR GTA: TRAP	(n = 65) PCOS (IVF) (n = 98) Non-PCOS (IVF)	30.3 ± 4.3 31.7 ± 3.8	GTL: shorter in PCOS GTA: no difference Earlier infertility symptoms when shorter TL and lower TA
Wei et al., 2017 [75]	GTL: PCR	(n = 75) PCOS (IVF) (n = 81) Non-PCOS (IVF)	28.36 ± 2.55 28.09 ± 2.26	GTL: longer in PCOS
Pedroso et al., 2020 [82]	GTL: PCR	(n = 43) PCOS (IVF) (n = 67) Non-PCOS (IVF)	33.7 ± 4.1	GTL: no difference GTA: higher in PCOS Similar TA and TL in GV and MI oocytes

GTL, granulosa telomere length; GTA, granulosa telomerase activity; IVF: *in vitro* fertilization; GV, germinative vesicle; MI, oocyte at the metaphase of the first meiosis division; TRAP, telomere repeat amplification protocol; PCR, polymerase chain reaction.

### 5. Premature Ovarian Failure

Premature ovarian failure (POF) is a clinical syndrome defined by loss of ovarian function before the age of 40 years. Its prevalence is about 1% [87]. Symptoms include menstrual disturbance (oligo or amenorrhea) with raised gonadotrophins and decreased estradiol. As a diagnosis criterion, the European Society of Human Reproduction and Embryology (ESHRE) working group has suggested an oligo-amenorrhea of at least 4 months and FSH >25 IU/L twice, 4 weeks apart [88]. Currently, spontaneous pregnancy rates in women with idiopathic POF are low (between 5 and 10%), and there is no treatment to improve the chances of spontaneous conception [89]. The long-term consequences of POF are the same as menopause and include osteoporosis, cognitive dysfunction, urogenital symptoms, such as vaginal dryness, increased risk of cardiovascular and autoimmune disease, as well as all-cause mortality [90]. The symptoms and long-term consequences of POF are summarized in Table 3.

Short GTL and/or low GTA could limit the mitosis ability of granulosa cells and, therefore, impair oocyte competence, resulting in infertility since granulosa cells must divide intensely during follicular maturation [91–93].

**Table 3.** POF symptoms and long-term conditions [93].

	Clinic and Biologic Symptoms	Long-Term Clinical Consequences
	Ovarian dysfunction (Oligo-amenorrhea for at least 4 months)	Infertility
Diagnostic criteria according to ESHRE	Clinical estrogen deficiency (climacteric syndrome) and sex hormonal dysfunction with: elevated FSH level >25 IU/L on two occasions >4 weeks apart	Urogenital symptoms (vaginal dryness, irritation and itching, sexual disorders) Cognitive dysfunction (memory and concentration problems, increased risk of dementia) Autoimmunity (increased risk of autoimmune disease) Bone alteration (osteopenia, osteoporosis, and increased risk of fracture) Increased all-cause mortality

Several studies have focused on the potential correlation between telomere length and/or telomerase activity in granulosa cells in women with POF. Some authors have hypothesized the existence of a possible link between LTL and female fertility since TL are synchronized in somatic tissues at birth and that strong correlations subsist later in life [94].



The confirmation of their hypothesis would lead to a POF predictive marker, more easily available than GTL. Unfortunately, the results are contradictory [95–97].

Looking for an explanation for these contradictory results, some authors have investigated telomerase activity in ovaries: In 2000, Kinugawa et al. [98] demonstrated that telomerase is active in ovaries and its activity normally decreased with age and in POF patients with follicle depletion. Butts et al. [60] and Xu et al. [99] found shorter telomeres and a lower TA in the granulosa cells of POF women than in healthy controls. These studies suggest that short telomeres could be associated with a fertility decrease and a shorter window of reproductive lifespan [60,99]. The hypothesis is that short telomeres could limit the proliferation capacity of granulosa cells and, in doing so, induce POF. Moreover, lower telomerase activity in granulosa cells would compensate less for telomeric attrition, leading to a limitation in the cell proliferation required for correct follicle maturation [48,60,98,100].

In conclusion, shorter telomeres and lower telomerase activity in granulosa cells seem to be associated with premature ovarian failure. The results are shown in Table 4.

**Table 4.** Studies associating telomere length and telomerase activity in granulosa cells/ovaries with POF.

Authors	Method	Population	Population Age (Years)	Results
Kinugawa et al., 2000 [98]	TRAP	(n = 5) POF (n = 20) Controls	31.4 <38	Ovarian TA: lower level in POF
Butts et al., 2009 [60]	PCR TRAP	(n = 12) POF (n = 42) Controls	30–37 23–37	GTL: shorter in POF GTA: absence of TA is more frequent in POF
Xu et al., 2017 [99]	PCR TRAP	(n = 120) POF (n = 90) Controls	32.95 ± 4.27 29.98 ± 4.28	GTL: shorter in POF GTA: lower level in POF

GTL, granulosa telomere length; GTA, granulosa telomerase activity; POF, premature ovarian failure; TRAP, telomere repeat amplification protocol; PCR, polymerase chain reaction.

These parameters could be an early and reliable marker of the decline in ovarian function. However, these results should be confirmed with larger studies with more subjects and the most suitable method [93]. The extrapolation of leukocyte telomere length does not seem possible, perhaps because initial telomere length and telomerase activity in granulosa cells can be impacted by some acquired factors such as in situ or general oxidative stress.

## 6. Discussion

As commonly agreed, telomere length/telomerase activity is a crucial factor in the proliferation capacity of cells in a tissue. Gonadal tissues present specificities in both sexes; in men, telomerase is constitutively activated throughout life in spermatogonia, whereas in women, TA partially compensating for telomere shortening declines with age in ovaries. “Ovarian aging” is, therefore, greatly reliant on TA in granulosa cells.

Based on this hypothesis, the ovarian reserve of follicles appears the result of a complex function in which the follicle number at a time “t” depends on the pool of follicles established at the fifth month of intrauterine life, with the cumulative number of follicles disappearing monthly until the time “t.” If the pool of primordial follicles established during the first months of pregnancy seems more reliable to ovogonial telomere length and ovogonial telomerase activity, the kinetics of disappearance of follicles after birth seems dependent on the telomere length/telomerase activity in granulosa cells. Mathematical models were used to predict infertility with aging according to TL and TA in granulosa cells. Predicted infertility was associated with either low levels of telomerase activity and short telomeres in GCs or with cumulative exposure to oxidative stress [101,102]. The conclusion of these models is the important role of sustained ovarian TA to preserve fertility in aging. Interestingly, it was shown that telomerase activity in luteinized granulosa cells was positively associated with pregnancy rate during *in vitro* fertilization (IVF)

treatment [103]. Moreover, *TERT* SNPs are also associated with IVF outcomes [104]. In Dyskeratosis congenita (DKC), canonical human telomeropathy is characterized by low telomerase activity and short telomeres, and women display diminished ovarian reserves, as estimated by circulating levels of anti-Mullerian hormone [105] and impaired reproductive function [106]. These results are consistent with the hypothesis of granulosa telomerase activity as a potential mechanism to preserve fertility.

Although this theory appears confirmed by the studies of GTL and GTA in the case of POF, the results of studies provided in PCOS populations appear controversial. In PCOS, the initial ovarian reserve of primordial follicles is abnormally rich, but the proliferation capacity of granulosa cells is sometimes high, explaining the high rate of twoness in PCOS populations and an increased risk of ovarian cancer [107]. In contrast, the proliferation capacity of granulosa cells is probably sometimes diminished, explaining the high rate of small follicles with immature oocytes picked up after ovarian stimulation in the context of IVF. The variable expression of GTA could be explained by two major types of factors: on the one hand, high levels of estrogen and androgen, which increase telomerase activity via stimulation of the *TERT* promoter, and on the other hand, some factors such as oxidative stress, which are generated at a high level in PCOS patients, impair telomere length homeostasis or lead the cells to enter apoptosis via a non-canonical pathway, implying mitochondria dysfunction.

The impact of oxidative stress is probably major in cases of inflammatory disease of ovaries such as endometriosis with ovarian implants of endometrial tissue. In endometriosis, endometrial telomerase is abnormally activated because of the endogenous autonomous high estrogen secretion of endometrial cells linked to a constitutive abnormally stimulated aromatase [108]. This estrogen source stimulates the telomerase activity of endometrial cells, explaining the invasive and proliferative characteristics of these cells colonizing ovaries and targeting the inflammatory state at the origin of major oxidative stress. This oxidative stress strongly decreases TA in GCs [81] via NF- $\kappa$ B pathways, finally impairing oocyte quality. Li et al. proposed that intrafollicular TNF- $\alpha$  might also downregulate TA and *TERT* expression via the NF- $\kappa$ B pathway. Thus, special attention should be given to limit/avoid any situation leading to important oxidative stress in the ovaries.

As proposed by some authors, restoring ovarian activity could be obtained by inducing the apparition of new stem cells, inducing pluripotent stem cells (IPS) able to differentiate into oocytes, or stimulating the proliferation of existing ovarian cells [109] without increasing the risk of oncogenesis. Among the strategies to reactivate telomerase, the exposition of ovaries to four factors, identified by Yamanaka and Takahashi (Oct3/4, Sox2, c-Myc, and Klf4) [110], is often cited [109] since it appears sufficient to induce IPS. As presented above, c-Myc and Klf4 are known activators of TA [111,112] and could also reactivate dormant tissular stem cells. Animal experiments on telomerase activation have succeeded in increasing the longevity of mice without increasing the development of tumors [113]. Moreover, in aged telomerase-deficient mice, telomerase reactivation reversed tissue degeneration and allowed the restoration of animal fertility [114]. In humans, various regenerative assays have been provided. In some of them, platelet-rich plasma (PRP) has been used, restoring the proliferation capacity of dormant stem cells [115]. In some clinical trials, these therapeutics were implemented into routine clinical practice as a rejuvenating agent or to promote healing in dermatology, plastic surgery, dentistry, orthopedics, and, in a similar way, gynecology [116] in order to optimize IVF outcomes in POF patients. PRP contains many growth factors supporting the survey and proliferation of ovarian stem cells directly or indirectly through the activation of telomerase [117]. Nevertheless, by activating telomerase using either a genic therapy strategy or treatment, the risk of cancer seems a real threat [118].

Indeed, telomerase activity is found in all types of ovarian cancers [119,120]. Similar to the majority of telomerase-positive cancers, ovarian cancers display short TL and high TA [119]. Ovarian cancers originate from ovarian surface epithelial cells [121]. These cells are telomerase negative [120]. Telomerase activation is an early event in ovarian carcino-

genesis [57], and the association of TERT expression with inactivation of p53 and activation of the Ras, Bcl2, and c-myc pathways are critical in oncogenic transformation [121]. Interestingly, these genes are estrogen-responsive genes [122,123]. Ovarian carcinogenesis could originate from estrogen paracrine activity. Estrogen from the granulosa cell compartment could reach the epithelial cells, inducing telomerase activation and oncogenic transformation of the ovarian epithelium [124]. The levels of telomerase activity correlate with the severity and recurrence of ovarian cancers [125]. Recently, effective telomerase-molecular targeted therapy has been tested as an approach for the treatment of ovarian cancer [126].

The presence of the *TERT* promoter in a dense CG-rich CpG island suggests a role for methylation in the regulation of TERT expression [127]. In cows, an age-dependent decrease in the global methylation stage correlates with a decrease in telomerase activity and a decrease in the proliferation capacity of granulosa cells [55].

Recently, some dietary recommendations have been proposed to improve telomerase activity [128] through their antioxidative role and impact on promoter methylation. However, investigations to understand the expression and methylation pattern of TERT in human ovaries [129] do not agree with animal studies; the methylation levels of the human *TERT* promoter do not seem to correlate with the level of TA. Therefore, all dietary recommendations, such as the Mediterranean diet, show that nutrient-promoting DNA methylations have probably more impact on TA through their antioxidative role than through their ability to improve ovarian TA. Similarly, the last decade has seen a great of studies analyzing the possible relation between the Mediterranean diet and TL. Most of the studies showed an association of the Mediterranean diet with longer telomeres [130,131]. However, these were almost exclusively cross-sectional and correlational studies, making causal interpretation problematic [132]. Longitudinal studies and randomized controlled trials are very limited, and their results are inconclusive [133,134]. Therefore, dietary recommendations may have an effect on ovarian aging without a direct effect on TA activity or TL, probably through oxidative stress modulation.

During adult life, in addition to their proliferative role in granulosa cells, TERT and TERC seem to globally protect ovarian mitochondria from oxidative stress [35,36,41], indirectly promoting mitochondrial steroidogenesis and, especially, estrogen synthesis. A non-proliferative role of both subunits of telomerase appears similar to a complementary amplification loop of its proliferative function since estrogen upregulates TERT expression [135]. Furthermore, a similar non-proliferative role of telomerase subunits may directly protect oocytes through their mitochondria, playing a role in oocyte competence, especially with aging [136]. Age-dependent mtDNA instability and the accumulation of mtDNA mutations in the oocyte result in the alteration of oocyte competence. Mitochondrial TERT may protect oocytes from these effects by binding to mtDNA.

## 7. Conclusions (Abstract Figure)

In the field of female fertility, telomerase presents a special interest since the capacity for cell proliferation is at the heart of embryonic development, particularly in fetal oocyte proliferation and during adult life proliferation of granulosa cells. Classical telomerase proliferative activity is upregulated mainly by estrogens by activation of the promoter and downregulated by different agents of oxidative stress. Moreover, through their interaction with mitochondria, telomerase subunits can more subtly modulate oocyte competence. The hypothesis in which telomerase activity correlates with ovarian reserves of follicles and global female fertility appears confirmed by pathology, especially premature ovarian failure, which is characterized by a poor ovarian reserve and low TA. However, PCOS, which is associated with a rich ovarian reserve, does not always match with long granulosa telomere length and high ovarian TA, probably because of the implication of downstream regulators such as oxidative stress.

Improving ovarian TA has been explored as a very promising rejuvenation technique. Unfortunately, no activator agent is without risk of activation telomerase in dormant cancer cells. At best, we can hope to limit the loss of natural fertility or infertility linked

to a pathology by combating any factor jeopardizing the activity of telomerase. For this, hygiene of life and safe alimentation appear complementary to traditional strategies of fertility cryopreservation.

**Author Contributions:** Conceptualization, S.T., A.-J.F., and I.K.; writing—original draft preparation, S.T., A.-J.F., and I.K.; writing—review and editing, S.T., A.-J.F., S.N.T., A.B., J.-L.G., and I.K. S.T. and A.-J.F. contributed equally to this work. All authors have read and agreed to the published version of the manuscript.

**Funding:** This work was supported by the French PIA project Lorraine Université d’Excellence, which includes two University-Hospital Federation (FHU ARRIMAGE and CARTAGE), reference ANR-15-IDEX-04-LUE, the Investments for the Future program under grant agreement No ANR-15-RHU-0004.

**Institutional Review Board Statement:** Not applicable.

**Informed Consent Statement:** Not applicable.

**Conflicts of Interest:** The authors declare no conflict of interest.

## References

- López-Otín, C.; Blasco, M.A.; Partridge, L.; Serrano, M.; Kroemer, G. The Hallmarks of Aging. *Cell* **2013**, *153*, 1194–1217. [[CrossRef](#)]
- Greider, C.W.; Blackburn, E.H. Identification of a Specific Telomere Terminal Transferase Activity in Tetrahymena Extracts. *Cell* **1985**, *43*, 405–413. [[CrossRef](#)]
- Blackburn, E.H.; Collins, K. Telomerase: An RNP Enzyme Synthesizes DNA. *Cold Spring Harb. Perspect. Biol.* **2011**, *3*. [[CrossRef](#)] [[PubMed](#)]
- Wright, W.E.; Piatyszek, M.A.; Rainey, W.E.; Byrd, W.; Shay, J.W. Telomerase Activity in Human Germline and Embryonic Tissues and Cells. *Dev. Genet.* **1996**, *18*, 173–179. [[CrossRef](#)]
- Hiyama, E.; Hiyama, K. Telomere and Telomerase in Stem Cells. *Br. J. Cancer* **2007**, *96*, 1020–1024. [[CrossRef](#)] [[PubMed](#)]
- Shay, J.W.; Wright, W.E. Role of Telomeres and Telomerase in Cancer. *Semin. Cancer Biol.* **2011**, *21*, 349–353. [[CrossRef](#)] [[PubMed](#)]
- Weng, N.P.; Levine, B.L.; June, C.H.; Hodes, R.J. Regulated Expression of Telomerase Activity in Human T Lymphocyte Development and Activation. *J. Exp. Med.* **1996**, *183*, 2471–2479. [[CrossRef](#)]
- Shay, J.W.; Wright, W.E. Telomerase Activity in Human Cancer. *Curr. Opin. Oncol.* **1996**, *8*, 66–71. [[CrossRef](#)]
- Shay, J.W.; Wright, W.E. Telomeres and Telomerase: Implications for Cancer and Aging. *Radiat. Res.* **2001**, *155*, 188–193. [[CrossRef](#)]
- Shay, J.W.; Bacchetti, S. A Survey of Telomerase Activity in Human Cancer. *Eur. J. Cancer* **1997**, *33*, 787–791. [[CrossRef](#)]
- Feng, J.; Funk, W.D.; Wang, S.S.; Weinrich, S.L.; Avilion, A.A.; Chiu, C.P.; Adams, R.R.; Chang, E.; Allsopp, R.C.; Yu, J. The RNA Component of Human Telomerase. *Science* **1995**, *269*, 1236–1241. [[CrossRef](#)]
- Shay, J.W.; Wright, W.E. Telomeres and Telomerase: Three Decades of Progress. *Nat. Rev. Genet.* **2019**, *20*, 299–309. [[CrossRef](#)]
- Weinrich, S.L.; Pruzan, R.; Ma, L.; Ouellette, M.; Tesmer, V.M.; Holt, S.E.; Bodnar, A.G.; Lichtsteiner, S.; Kim, N.W.; Trager, J.B.; et al. Reconstitution of Human Telomerase with the Template RNA Component HTR and the Catalytic Protein Subunit HTRT. *Nat. Genet.* **1997**, *17*, 498–502. [[CrossRef](#)]
- Wu, R.A.; Upton, H.E.; Vogan, J.M.; Collins, K. Telomerase Mechanism of Telomere Synthesis. *Annu. Rev. Biochem.* **2017**, *86*, 439–460. [[CrossRef](#)]
- Mitchell, J.R.; Wood, E.; Collins, K. A Telomerase Component Is Defective in the Human Disease Dyskeratosis Congenita. *Nature* **1999**, *402*, 551–555. [[CrossRef](#)]
- Egan, E.D.; Collins, K. Biogenesis of Telomerase Ribonucleoproteins. *RNA* **2012**, *18*, 1747–1759. [[CrossRef](#)] [[PubMed](#)]
- Holt, S.E.; Aisner, D.L.; Baur, J.; Tesmer, V.M.; Dy, M.; Ouellette, M.; Trager, J.B.; Morin, G.B.; Toft, D.O.; Shay, J.W.; et al. Functional Requirement of P23 and Hsp90 in Telomerase Complexes. *Genes Dev.* **1999**, *13*, 817–826. [[CrossRef](#)] [[PubMed](#)]
- Venteicher, A.S.; Abreu, E.B.; Meng, Z.; McCann, K.E.; Terns, R.M.; Veenstra, T.D.; Terns, M.P.; Artandi, S.E. A Human Telomerase Holoenzyme Protein Required for Cajal Body Localization and Telomere Synthesis. *Science* **2009**, *323*, 644–648. [[CrossRef](#)] [[PubMed](#)]
- Trinkle-Mulcahy, L.; Sleeman, J.E. The Cajal Body and the Nucleolus: “In a Relationship” or “It’s Complicated”? *RNA Biol.* **2017**, *14*, 739–751. [[CrossRef](#)] [[PubMed](#)]
- Tomlinson, R.L.; Ziegler, T.D.; Supakorndej, T.; Terns, R.M.; Terns, M.P. Cell Cycle-Regulated Trafficking of Human Telomerase to Telomeres. *Mol. Biol. Cell* **2006**, *17*, 955–965. [[CrossRef](#)] [[PubMed](#)]
- Wu, P.; De Lange, T. Human Telomerase Caught in the Act. *Cell* **2009**, *138*, 432–434. [[CrossRef](#)]
- Zhao, Y.; Sfeir, A.J.; Zou, Y.; Buseman, C.M.; Chow, T.T.; Shay, J.W.; Wright, W.E. Telomere Extension Occurs at Most Chromosome Ends and Is Uncoupled from Fill-in in Human Cancer Cells. *Cell* **2009**, *138*, 463–475. [[CrossRef](#)]
- Teixeira, M.T.; Arneric, M.; Sperisen, P.; Lingner, J. Telomere Length Homeostasis Is Achieved via a Switch between Telomerase-Extendible and -Nonextendible States. *Cell* **2004**, *117*, 323–335. [[CrossRef](#)]



24. Greider, C.W. Regulating Telomere Length from the inside out: The Replication Fork Model. *Genes Dev.* **2016**, *30*, 1483–1491. [[CrossRef](#)]
25. Ségal-Bendirdjian, E.; Geli, V. Non-Canonical Roles of Telomerase: Unraveling the Imbroglia. *Front. Cell Dev. Biol.* **2019**, *7*. [[CrossRef](#)] [[PubMed](#)]
26. Lindvall, C.; Hou, M.; Komurasaki, T.; Zheng, C.; Henriksson, M.; Sedivy, J.M.; Björkholm, M.; Teh, B.T.; Nordenskjöld, M.; Xu, D. Molecular Characterization of Human Telomerase Reverse Transcriptase-Immortalized Human Fibroblasts by Gene Expression Profiling: Activation of the Epiregulin Gene. *Cancer Res.* **2003**, *63*, 1743–1747.
27. Ghosh, A.; Saginc, G.; Leow, S.C.; Khattar, E.; Shin, E.M.; Yan, T.D.; Wong, M.; Zhang, Z.; Li, G.; Sung, W.-K.; et al. Telomerase Directly Regulates NF- $\kappa$ B-Dependent Transcription. *Nat. Cell Biol.* **2012**, *14*, 1270–1281. [[CrossRef](#)]
28. Park, J.-I.; Venteicher, A.S.; Hong, J.Y.; Choi, J.; Jun, S.; Shkrel, M.; Chang, W.; Meng, Z.; Cheung, P.; Ji, H.; et al. Telomerase Modulates Wnt Signalling by Association with Target Gene Chromatin. *Nature* **2009**, *460*, 66–72. [[CrossRef](#)] [[PubMed](#)]
29. Liu, Z.; Li, Q.; Li, K.; Chen, L.; Li, W.; Hou, M.; Liu, T.; Yang, J.; Lindvall, C.; Björkholm, M.; et al. Telomerase Reverse Transcriptase Promotes Epithelial-Mesenchymal Transition and Stem Cell-like Traits in Cancer Cells. *Oncogene* **2013**, *32*, 4203–4213. [[CrossRef](#)] [[PubMed](#)]
30. Zhou, L.; Zheng, D.; Wang, M.; Cong, Y.-S. Telomerase Reverse Transcriptase Activates the Expression of Vascular Endothelial Growth Factor Independent of Telomerase Activity. *Biochem. Biophys. Res. Commun.* **2009**, *386*, 739–743. [[CrossRef](#)]
31. Liu, N.; Ding, D.; Hao, W.; Yang, F.; Wu, X.; Wang, M.; Xu, X.; Ju, Z.; Liu, J.-P.; Song, Z.; et al. HTERT Promotes Tumor Angiogenesis by Activating VEGF via Interactions with the Sp1 Transcription Factor. *Nucleic Acids Res.* **2016**, *44*, 8693–8703. [[CrossRef](#)] [[PubMed](#)]
32. Yu, J.; Yuan, X.; Sjöholm, L.; Liu, T.; Kong, F.; Ekström, T.J.; Björkholm, M.; Xu, D. Telomerase Reverse Transcriptase Regulates DNMT3B Expression/Aberrant DNA Methylation Phenotype and AKT Activation in Hepatocellular Carcinoma. *Cancer Lett.* **2018**, *434*, 33–41. [[CrossRef](#)]
33. Young, J.I.; Sedivy, J.M.; Smith, J.R. Telomerase Expression in Normal Human Fibroblasts Stabilizes DNA 5-Methylcytosine Transferase, I. *J. Biol. Chem.* **2003**, *278*, 19904–19908. [[CrossRef](#)] [[PubMed](#)]
34. Santos, J.H.; Meyer, J.N.; Van Houten, B. Mitochondrial Localization of Telomerase as a Determinant for Hydrogen Peroxide-Induced Mitochondrial DNA Damage and Apoptosis. *Hum. Mol. Genet.* **2006**, *15*, 1757–1768. [[CrossRef](#)] [[PubMed](#)]
35. Ahmed, S.; Passos, J.F.; Birket, M.J.; Beckmann, T.; Brings, S.; Peters, H.; Birch-Machin, M.A.; von Zglinicki, T.; Saretzki, G. Telomerase Does Not Counteract Telomere Shortening but Protects Mitochondrial Function under Oxidative Stress. *J. Cell Sci.* **2008**, *121*, 1046–1053. [[CrossRef](#)] [[PubMed](#)]
36. Haendeler, J.; Dröse, S.; Büchner, N.; Jakob, S.; Altschmied, J.; Goy, C.; Spyridopoulos, I.; Zeiher, A.M.; Brandt, U.; Dimmeler, S. Mitochondrial Telomerase Reverse Transcriptase Binds to and Protects Mitochondrial DNA and Function from Damage. *Arterioscler. Thromb. Vasc. Biol.* **2009**, *29*, 929–935. [[CrossRef](#)]
37. Indran, I.R.; Hande, M.P.; Pervaiz, S. HTERT Overexpression Alleviates Intracellular ROS Production, Improves Mitochondrial Function, and Inhibits ROS-Mediated Apoptosis in Cancer Cells. *Cancer Res.* **2011**, *71*, 266–276. [[CrossRef](#)]
38. Santos, J.H.; Meyer, J.N.; Skovvaga, M.; Annab, L.A.; Houten, B.V. Mitochondrial HTERT Exacerbates Free-Radical-Mediated MtDNA Damage. *Aging Cell* **2004**, *3*, 399–411. [[CrossRef](#)]
39. Shin, W.H.; Chung, K.C. Human Telomerase Reverse Transcriptase Positively Regulates Mitophagy by Inhibiting the Processing and Cytoplasmic Release of Mitochondrial PINK1. *Cell Death Dis.* **2020**, *11*, 425. [[CrossRef](#)]
40. Cheng, Y.; Liu, P.; Zheng, Q.; Gao, G.; Yuan, J.; Wang, P.; Huang, J.; Xie, L.; Lu, X.; Tong, T.; et al. Mitochondrial Trafficking and Processing of Telomerase RNA TERC. *Cell Rep.* **2018**, *24*, 2589–2595. [[CrossRef](#)]
41. Zheng, Q.; Liu, P.; Gao, G.; Yuan, J.; Wang, P.; Huang, J.; Xie, L.; Lu, X.; Di, F.; Tong, T.; et al. Mitochondrion-Processed TERC Regulates Senescence without Affecting Telomerase Activities. *Protein Cell* **2019**, *10*, 631–648. [[CrossRef](#)] [[PubMed](#)]
42. Baker, T.G. A Quantitative And Cytological Study Of Germ Cells In Human Ovaries. *Proc. R. Soc. Lond. B Biol. Sci.* **1963**, *158*, 417–433. [[CrossRef](#)] [[PubMed](#)]
43. Pepling, M.E. Follicular Assembly: Mechanisms of Action. *Reprod. Camb. Engl.* **2012**, *143*, 139–149. [[CrossRef](#)]
44. Keefe, D.L.; Marquard, K.; Liu, L. The Telomere Theory of Reproductive Senescence in Women. *Curr. Opin. Obstet. Gynecol.* **2006**, *18*, 280–285. [[CrossRef](#)] [[PubMed](#)]
45. Gougeon, A. Ovarian Follicular Growth in Humans: Ovarian Ageing and Population of Growing Follicles. *Maturitas* **1998**, *30*, 137–142. [[CrossRef](#)]
46. Rodgers, R.J.; Irving-Rodgers, H.F. Morphological Classification of Bovine Ovarian Follicles. *Reprod. Camb. Engl.* **2010**, *139*, 309–318. [[CrossRef](#)]
47. Lavranos, T.C.; Mathis, J.M.; Latham, S.E.; Kalionis, B.; Shay, J.W.; Rodgers, R.J. Evidence for Ovarian Granulosa Stem Cells: Telomerase Activity and Localization of the Telomerase Ribonucleic Acid Component in Bovine Ovarian Follicles. *Biol. Reprod.* **1999**, *61*, 358–366. [[CrossRef](#)]
48. Kosebent, E.G.; Uysal, F.; Ozturk, S. Telomere Length and Telomerase Activity during Folliculogenesis in Mammals. *J. Reprod. Dev.* **2018**, *64*, 477–484. [[CrossRef](#)]
49. Clément, F.; Gruet, M.A.; Monget, P.; Terqui, M.; Jolivet, E.; Monniaux, D. Growth Kinetics of the Granulosa Cell Population in Ovarian Follicles: An Approach by Mathematical Modelling. *Cell Prolif.* **1997**, *30*, 255–270. [[CrossRef](#)]



50. Van Deerlin, P.G.; Cekleniak, N.; Coutifaris, C.; Boyd, J.; Strauss, J.F. Evidence for the Oligoclonal Origin of the Granulosa Cell Population of the Mature Human Follicle. *J. Clin. Endocrinol. Metab.* **1997**, *82*, 3019–3024. [[CrossRef](#)]
51. Kossowska-Tomaszczuk, K.; De Geyter, C. Cells with Stem Cell Characteristics in Somatic Compartments of the Ovary. *BioMed Res. Int.* **2013**, *2013*, 1–8. [[CrossRef](#)] [[PubMed](#)]
52. Slack, J.M.W. What Is a Stem Cell? *WIREs Dev. Biol.* **2018**, *7*, e323. [[CrossRef](#)] [[PubMed](#)]
53. Lavranos, T.C. Anchorage-Independent Culture of Bovine Granulosa Cells: The Effects of Basic Fibroblast Growth Factor and Dibutyryl CAMP on Cell Division and Differentiation. *Exp. Cell Res.* **1994**, *211*, 245–251. [[CrossRef](#)] [[PubMed](#)]
54. Kossowska-Tomaszczuk, K.; Geyter, C.D.; Geyter, M.D.; Martin, I.; Holzgreve, W.; Scherberich, A.; Zhang, H. The Multipotency of Luteinizing Granulosa Cells Collected from Mature Ovarian Follicles. *Stem Cells* **2009**, *27*, 210–219. [[CrossRef](#)]
55. Goto, H.; Iwata, H.; Takeo, S.; Nisinonso, K.; Murakami, S.; Monji, Y.; Kuwayama, T. Effect of Bovine Age on the Proliferative Activity, Global DNA Methylation, Relative Telomere Length and Telomerase Activity of Granulosa Cells. *Zygote Camb. Engl.* **2013**, *21*, 256–264. [[CrossRef](#)]
56. Russo, V.; Berardinelli, P.; Martelli, A.; Giacinto, O.D.; Nardinocchi, D.; Fantasia, D.; Barboni, B. Expression of Telomerase Reverse Transcriptase Subunit (TERT) and Telomere Sizing in Pig Ovarian Follicles. *J. Histochem. Cytochem.* **2006**, *54*, 443–455. [[CrossRef](#)]
57. Liu, J.-P.; Li, H. Telomerase in the Ovary. *Reprod. Camb. Engl.* **2010**, *140*, 215–222. [[CrossRef](#)]
58. Rodgers, R.J.; Irving-Rodgers, H.F.; van Wezel, I.L.; Krupa, M.; Lavranos, T.C. Dynamics of the Membrana Granulosa during Expansion of the Ovarian Follicular Antrum. *Mol. Cell. Endocrinol.* **2001**, *171*, 41–48. [[CrossRef](#)]
59. Tománek, M.; Chronowska, E.; Kott, T.; Czerneková, V. Telomerase Activity in Pig Granulosa Cells Proliferating and Differentiating in Vitro. *Anim. Reprod. Sci.* **2008**, *104*, 284–298. [[CrossRef](#)]
60. Butts, S.; Riethman, H.; Ratcliffe, S.; Shaunik, A.; Coutifaris, C.; Barnhart, K. Correlation of Telomere Length and Telomerase Activity with Occult Ovarian Insufficiency. *J. Clin. Endocrinol. Metab.* **2009**, *94*, 4835–4843. [[CrossRef](#)]
61. Kosebent, E.G.; Uysal, F.; Ozturk, S. The Altered Expression of Telomerase Components and Telomere-Linked Proteins May Associate with Ovarian Aging in Mouse. *Exp. Gerontol.* **2020**, *138*, 110975. [[CrossRef](#)]
62. Uysal, F.; Kosebent, E.G.; Toru, H.S.; Ozturk, S. Decreased Expression of TERT and Telomeric Proteins as Human Ovaries Age May Cause Telomere Shortening. *J. Assist. Reprod. Genet.* **2021**, *38*, 429–441. [[CrossRef](#)]
63. Gardner, M.; Bann, D.; Wiley, L.; Cooper, R.; Hardy, R.; Nitsch, D.; Martin-Ruiz, C.; Shiels, P.; Sayer, A.A.; Barbieri, M.; et al. Gender and Telomere Length: Systematic Review and Meta-Analysis. *Exp. Gerontol.* **2014**, *51*, 15–27. [[CrossRef](#)]
64. Mayer, S.; Brüderlein, S.; Perner, S.; Waibel, I.; Holdenried, A.; Ciloglu, N.; Hasel, C.; Mattfeldt, T.; Nielsen, K.V.; Möller, P. Sex-Specific Telomere Length Profiles and Age-Dependent Erosion Dynamics of Individual Chromosome Arms in Humans. *Cytogenet. Genome Res.* **2006**, *112*, 194–201. [[CrossRef](#)]
65. Lulkiewicz, M.; Bajsert, J.; Kopczynski, P.; Barczak, W.; Rubis, B. Telomere Length: How the Length Makes a Difference. *Mol. Biol. Rep.* **2020**, *47*, 7181–7188. [[CrossRef](#)]
66. Chen, W.; Kimura, M.; Kim, S.; Cao, X.; Srinivasan, S.R.; Berenson, G.S.; Kark, J.D.; Aviv, A. Longitudinal versus Cross-Sectional Evaluations of Leukocyte Telomere Length Dynamics: Age-Dependent Telomere Shortening Is the Rule. *J. Gerontol. A Biol. Sci. Med. Sci.* **2011**, *66*, 312–319. [[CrossRef](#)] [[PubMed](#)]
67. Dalgård, C.; Benetos, A.; Verhulst, S.; Labat, C.; Kark, J.D.; Christensen, K.; Kimura, M.; Kyvik, K.O.; Aviv, A. Leukocyte Telomere Length Dynamics in Women and Men: Menopause vs. Age Effects. *Int. J. Epidemiol.* **2015**, *44*, 1688–1695. [[CrossRef](#)] [[PubMed](#)]
68. Factor-Litvak, P.; Susser, E.; Kezios, K.; McKeague, I.; Kark, J.D.; Hoffman, M.; Kimura, M.; Wapner, R.; Aviv, A. Leukocyte Telomere Length in Newborns: Implications for the Role of Telomeres in Human Disease. *Pediatrics* **2016**, *137*. [[CrossRef](#)] [[PubMed](#)]
69. Piontkewitz, Y.; Sundfeldt, K.; Hedin, L. The Expression of C-Myc during Follicular Growth and Luteal Formation in the Rat Ovary in Vivo. *J. Endocrinol.* **1997**, *152*, 395–406. [[CrossRef](#)] [[PubMed](#)]
70. Bayne, S.; Jones, M.E.E.; Li, H.; Liu, J.-P. Potential Roles for Estrogen Regulation of Telomerase Activity in Aging. *Ann. N. Y. Acad. Sci.* **2007**, *1114*, 48–55. [[CrossRef](#)] [[PubMed](#)]
71. Viña, J.; Borrás, C.; Gambini, J.; Sastre, J.; Pallardó, F.V. Why Females Live Longer than Males? Importance of the Upregulation of Longevity-Associated Genes by Oestrogenic Compounds. *FEBS Lett.* **2005**, *579*, 2541–2545. [[CrossRef](#)]
72. Yamagata, Y.; Nakamura, Y.; Umayahara, K.; Harada, A.; Takayama, H.; Sugino, N.; Kato, H. Changes in Telomerase Activity in Experimentally Induced Atretic Follicles of Immature Rats. *Endocr. J.* **2002**, *49*, 589–595. [[CrossRef](#)]
73. Bayne, S.; Li, H.; Jones, M.E.E.; Pinto, A.R.; van Sinderen, M.; Drummond, A.; Simpson, E.R.; Liu, J.-P. Estrogen Deficiency Reversibly Induces Telomere Shortening in Mouse Granulosa Cells and Ovarian Aging in Vivo. *Protein Cell* **2011**, *2*, 333–346. [[CrossRef](#)] [[PubMed](#)]
74. Mordechai, A.; Wasserman, M.; Abramov, M.; Ben-Menahem, D.; Har-Vardi, I.; Levitas, E.; Priel, E. Increasing Telomerase Enhanced Steroidogenic Genes Expression and Steroid Hormones Production in Rat and Human Granulosa Cells and in Mouse Ovary. *J. Steroid Biochem. Mol. Biol.* **2020**, *197*, 105551. [[CrossRef](#)] [[PubMed](#)]
75. Wei, D.; Xie, J.; Yin, B.; Hao, H.; Song, X.; Liu, Q.; Zhang, C.; Sun, Y. Significantly Lengthened Telomere in Granulosa Cells from Women with Polycystic Ovarian Syndrome (PCOS). *J. Assist. Reprod. Genet.* **2017**, *34*, 861–866. [[CrossRef](#)]
76. Dunaif, A. Insulin Resistance and the Polycystic Ovary Syndrome: Mechanism and Implications for Pathogenesis. *Endocr. Rev.* **1997**, *18*, 774–800. [[CrossRef](#)]

77. Ajmal, N.; Khan, S.Z.; Shaikh, R. Polycystic Ovary Syndrome (PCOS) and Genetic Predisposition: A Review Article. *Eur. J. Obstet. Gynecol. Reprod. Biol. X* **2019**, *3*. [[CrossRef](#)] [[PubMed](#)]
78. Li, Q.; Du, J.; Feng, R.; Xu, Y.; Wang, H.; Sang, Q.; Xing, Q.; Zhao, X.; Jin, L.; He, L.; et al. A Possible New Mechanism in the Pathophysiology of Polycystic Ovary Syndrome (PCOS): The Discovery That Leukocyte Telomere Length Is Strongly Associated with PCOS. *J. Clin. Endocrinol. Metab.* **2014**, *99*, E234–E240. [[CrossRef](#)] [[PubMed](#)]
79. Mohammadi, M. Oxidative Stress and Polycystic Ovary Syndrome: A Brief Review. *Int. J. Prev. Med.* **2019**, *10*, 86. [[CrossRef](#)] [[PubMed](#)]
80. Pedroso, D.C.C.; Miranda-Furtado, C.L.; Kogure, G.S.; Meola, J.; Okuka, M.; Silva, C.; Calado, R.T.; Ferriani, R.A.; Keefe, D.L.; dos Reis, R.M. Inflammatory Biomarkers and Telomere Length in Women with Polycystic Ovary Syndrome. *Fertil. Steril.* **2015**, *103*, 542–547.e2. [[CrossRef](#)] [[PubMed](#)]
81. Li, Y.; Li, R.; Ouyang, N.; Dai, K.; Yuan, P.; Zheng, L.; Wang, W. Investigating the Impact of Local Inflammation on Granulosa Cells and Follicular Development in Women with Ovarian Endometriosis. *Fertil. Steril.* **2019**, *112*, 882–891.e1. [[CrossRef](#)]
82. Pedroso, D.C.C.; Santana, V.P.; Donaires, F.S.; Picinato, M.C.; Giorgenon, R.C.; Santana, B.A.; Pimentel, R.N.; Keefe, D.L.; Calado, R.T.; Ferriani, R.A.; et al. Telomere Length and Telomerase Activity in Immature Oocytes and Cumulus Cells of Women with Polycystic Ovary Syndrome. *Reprod. Sci. Thousand Oaks Calif.* **2020**, *27*, 1293–1303. [[CrossRef](#)] [[PubMed](#)]
83. Vasilopoulos, E.; Fragkiadakis, P.; Kalliora, C.; Fragou, D.; Docea, A.O.; Vakonaki, E.; Tsoukalas, D.; Calina, D.; Buga, A.M.; Georgiadis, G.; et al. The Association of Female and Male Infertility with Telomere Length (Review). *Int. J. Mol. Med.* **2019**, *44*, 375–389. [[CrossRef](#)] [[PubMed](#)]
84. Calado, R.T.; Yewdell, W.T.; Wilkerson, K.L.; Regal, J.A.; Kajigaya, S.; Stratakis, C.A.; Young, N.S. Sex Hormones, Acting on the TERT Gene, Increase Telomerase Activity in Human Primary Hematopoietic Cells. *Blood* **2009**, *114*, 2236–2243. [[CrossRef](#)] [[PubMed](#)]
85. Li, Y.; Deng, B.; Ouyang, N.; Yuan, P.; Zheng, L.; Wang, W. Telomere Length Is Short in PCOS and Oral Contraceptive Does Not Affect the Telomerase Activity in Granulosa Cells of Patients with PCOS. *J. Assist. Reprod. Genet.* **2017**, *34*, 849–859. [[CrossRef](#)] [[PubMed](#)]
86. Webber, L.J.; Stubbs, S.; Stark, J.; Trew, G.H.; Margara, R.; Hardy, K.; Franks, S. Formation and Early Development of Follicles in the Polycystic Ovary. *Lancet Lond. Engl.* **2003**, *362*, 1017–1021. [[CrossRef](#)]
87. Beck-Peccoz, P.; Persani, L. Premature Ovarian Failure. *Orphanet. J. Rare Dis.* **2006**, *1*, 9. [[CrossRef](#)]
88. European Society for Human Reproduction and Embryology (ESHRE) Guideline Group on POI; Webber, L.; Davies, M.; Anderson, R.; Bartlett, J.; Braat, D.; Cartwright, B.; Cifkova, R.; de Muinck Keizer-Schrama, S.; Hogervorst, E.; et al. ESHRE Guideline: Management of Women with Premature Ovarian Insufficiency. *Hum. Reprod. Oxf. Engl.* **2016**, *31*, 926–937. [[CrossRef](#)]
89. Committee on Gynecologic Practice Committee Opinion No. 605: Primary Ovarian Insufficiency in Adolescents and Young Women. *Obstet. Gynecol.* **2014**, *124*, 193–197. [[CrossRef](#)]
90. Podfigurna-Stopa, A.; Czyzyk, A.; Grymowicz, M.; Smolarczyk, R.; Katulski, K.; Czajkowski, K.; Meczekalski, B. Premature Ovarian Insufficiency: The Context of Long-Term Effects. *J. Endocrinol. Invest.* **2016**, *39*, 983–990. [[CrossRef](#)]
91. Rocca, M.S.; Foresta, C.; Ferlin, A. Telomere Length: Lights and Shadows on Their Role in Human Reproduction. *Biol. Reprod.* **2019**, *100*, 305–317. [[CrossRef](#)] [[PubMed](#)]
92. Ozturk, S.; Sozen, B.; Demir, N. Telomere Length and Telomerase Activity during Oocyte Maturation and Early Embryo Development in Mammalian Species. *Mol. Hum. Reprod.* **2014**, *20*, 15–30. [[CrossRef](#)] [[PubMed](#)]
93. Fattet, A.-J.; Toupance, S.; Thornton, S.N.; Monnin, N.; Guéant, J.-L.; Benetos, A.; Kosciński, I. Telomere Length in Granulosa Cells and Leukocytes: A Potential Marker of Female Fertility? A Systematic Review of the Literature. *J. Ovarian Res.* **2020**, *13*, 96. [[CrossRef](#)] [[PubMed](#)]
94. Daniali, L.; Benetos, A.; Susser, E.; Kark, J.D.; Labat, C.; Kimura, M.; Desai, K.; Granick, M.; Aviv, A. Telomeres Shorten at Equivalent Rates in Somatic Tissues of Adults. *Nat. Commun.* **2013**, *4*, 1597. [[CrossRef](#)] [[PubMed](#)]
95. Hanna, C.W.; Bretherick, K.L.; Gair, J.L.; Fluker, M.R.; Stephenson, M.D.; Robinson, W.P. Telomere Length and Reproductive Aging. *Hum. Reprod. Oxf. Engl.* **2009**, *24*, 1206–1211. [[CrossRef](#)]
96. Miranda-Furtado, C.L.; Luchiari, H.R.; Chielli Pedroso, D.C.; Kogure, G.S.; Caetano, L.C.; Santana, B.A.; Santana, V.P.; Benetti-Pinto, C.L.; Reis, F.M.; Maciel, M.A.; et al. Skewed X-Chromosome Inactivation and Shorter Telomeres Associate with Idiopathic Premature Ovarian Insufficiency. *Fertil. Steril.* **2018**, *110*, 476–485.e1. [[CrossRef](#)] [[PubMed](#)]
97. Sayban, S.; Mirfakhraie, R.; Omrani, M.D.; Ghaedi, H.; Heidary, H.; Yaghoobi, H.; Azizi, F.; Pouresmaeili, F. Idiopathic Premature Ovarian Failure and Its Association to the Abnormal Longitudinal Changes of Telomere Length in a Population of Iranian Infertile Women: A Pilot Study. *Meta Gene* **2018**, *18*, 58–61. [[CrossRef](#)]
98. Kinugawa, C.; Murakami, T.; Okamura, K.; Yajima, A. Telomerase Activity in Normal Ovaries and Premature Ovarian Failure. *Tohoku J. Exp. Med.* **2000**, *190*, 231–238. [[CrossRef](#)]
99. Xu, X.; Chen, X.; Zhang, X.; Liu, Y.; Wang, Z.; Wang, P.; Du, Y.; Qin, Y.; Chen, Z.-J. Impaired Telomere Length and Telomerase Activity in Peripheral Blood Leukocytes and Granulosa Cells in Patients with Biochemical Primary Ovarian Insufficiency. *Hum. Reprod. Oxf. Engl.* **2017**, *32*, 201–207. [[CrossRef](#)]
100. Liu, C.C.; Ma, D.L.; Yan, T.-D.; Fan, X.; Poon, Z.; Poon, L.-F.; Goh, S.-A.; Rozen, S.G.; Hwang, W.Y.K.; Tergaonkar, V.; et al. Distinct Responses of Stem Cells to Telomere Uncapping—A Potential Strategy to Improve the Safety of Cell Therapy. *Stem Cells* **2016**, *34*, 2471–2484. [[CrossRef](#)]

101. Portillo, A.M.; Varela, E.; García-Velasco, J.A. Mathematical Model to Study the Aging of the Human Follicle According to the Telomerase Activity. *J. Theor. Biol.* **2019**, *462*, 446–454. [[CrossRef](#)]
102. Portillo, A.M.; Peláez, C. Mathematical Modelling of Ageing Acceleration of the Human Follicle Due to Oxidative Stress and Other Factors. *Math. Med. Biol. J. IMA* **2021**. [[CrossRef](#)]
103. Chen, H.; Wang, W.; Mo, Y.; Ma, Y.; Ouyang, N.; Li, R.; Mai, M.; He, Y.; Bodombossou-Djobo, M.M.A.; Yang, D. Women with High Telomerase Activity in Luteinised Granulosa Cells Have a Higher Pregnancy Rate during in Vitro Fertilisation Treatment. *J. Assist. Reprod. Genet.* **2011**, *28*, 797–807. [[CrossRef](#)] [[PubMed](#)]
104. Dai, K.; Xu, H.; Ouyang, N.; Li, Y.; Yuan, P.; Wang, L.; Zhao, X.; Wang, W. Correlation of Human Telomerase Reverse Transcriptase Single Nucleotide Polymorphisms with in Vitro Fertilisation Outcomes. *J. Assist. Reprod. Genet.* **2019**, *36*, 517–527. [[CrossRef](#)] [[PubMed](#)]
105. Sklavos, M.M.; Stratton, P.; Giri, N.; Alter, B.P.; Savage, S.A.; Pinto, L.A. Reduced Serum Levels of Anti-Müllerian Hormone in Females with Inherited Bone Marrow Failure Syndromes. *J. Clin. Endocrinol. Metab.* **2015**, *100*, E197–E203. [[CrossRef](#)]
106. Robinson, L.G.; Pimentel, R.; Wang, F.; Kramer, Y.G.; Gonullu, D.C.; Agarwal, S.; Navarro, P.A.; McCulloh, D.; Keefe, D.L. Impaired Reproductive Function and Fertility Preservation in a Woman with a Dyskeratosis Congenita. *J. Assist. Reprod. Genet.* **2020**, *37*, 1221–1225. [[CrossRef](#)] [[PubMed](#)]
107. Barry, J.A.; Azizia, M.M.; Hardiman, P.J. Risk of Endometrial, Ovarian and Breast Cancer in Women with Polycystic Ovary Syndrome: A Systematic Review and Meta-Analysis. *Hum. Reprod. Update* **2014**, *20*, 748–758. [[CrossRef](#)] [[PubMed](#)]
108. Hapangama, D.K.; Turner, M.A.; Drury, J.A.; Quenby, S.; Saretzki, G.; Martin-Ruiz, C.; Von Zglinicki, T. Endometriosis Is Associated with Aberrant Endometrial Expression of Telomerase and Increased Telomere Length. *Hum. Reprod. Oxf. Engl.* **2008**, *23*, 1511–1519. [[CrossRef](#)]
109. Polonio, A.M.; Chico-Sordo, L.; Córdova-Oriz, I.; Medrano, M.; García-Velasco, J.A.; Varela, E. Impact of Ovarian Aging in Reproduction: From Telomeres and Mice Models to Ovarian Rejuvenation. *Yale J. Biol. Med.* **2020**, *93*, 561–569.
110. Takahashi, K.; Yamanaka, S. Induction of Pluripotent Stem Cells from Mouse Embryonic and Adult Fibroblast Cultures by Defined Factors. *Cell* **2006**, *126*, 663–676. [[CrossRef](#)]
111. Wang, J.; Xie, L.Y.; Allan, S.; Beach, D.; Hannon, G.J. Myc Activates Telomerase. *Genes Dev.* **1998**, *12*, 1769–1774. [[CrossRef](#)]
112. Wong, C.-W.; Hou, P.-S.; Tseng, S.-F.; Chien, C.-L.; Wu, K.-J.; Chen, H.-F.; Ho, H.-N.; Kyo, S.; Teng, S.-C. Krüppel-like Transcription Factor 4 Contributes to Maintenance of Telomerase Activity in Stem Cells. *Stem Cells* **2010**, *28*, 1510–1517. [[CrossRef](#)]
113. Bernardes de Jesus, B.; Vera, E.; Schneeberger, K.; Tejera, A.M.; Ayuso, E.; Bosch, F.; Blasco, M.A. Telomerase Gene Therapy in Adult and Old Mice Delays Aging and Increases Longevity without Increasing Cancer. *EMBO Mol. Med.* **2012**, *4*, 691–704. [[CrossRef](#)]
114. Jaskelioff, M.; Muller, F.L.; Paik, J.-H.; Thomas, E.; Jiang, S.; Adams, A.C.; Sahin, E.; Kost-Alimova, M.; Protopopov, A.; Cadiñanos, J.; et al. Telomerase Reactivation Reverses Tissue Degeneration in Aged Telomerase-Deficient Mice. *Nature* **2011**, *469*, 102–106. [[CrossRef](#)]
115. Bretschneider, H.; Quade, M.; Lode, A.; Gelinsky, M.; Rammelt, S.; Zwingenberger, S.; Schaser, K.-D.; Vater, C. Characterization of Naturally Occurring Bioactive Factor Mixtures for Bone Regeneration. *Int. J. Mol. Sci.* **2020**, *21*, 1412. [[CrossRef](#)] [[PubMed](#)]
116. Cakiroglu, Y.; Saltik, A.; Yuceturk, A.; Karaosmanoglu, O.; Kopuk, S.Y.; Scott, R.T.; Tiras, B.; Seli, E. Effects of Intraovarian Injection of Autologous Platelet Rich Plasma on Ovarian Reserve and IVF Outcome Parameters in Women with Primary Ovarian Insufficiency. *Aging* **2020**, *12*, 10211–10222. [[CrossRef](#)]
117. Sills, E.S.; Wood, S.H. Autologous Activated Platelet-Rich Plasma Injection into Adult Human Ovary Tissue: Molecular Mechanism, Analysis, and Discussion of Reproductive Response. *Biosci. Rep.* **2019**, *39*. [[CrossRef](#)]
118. Shingu, T.; Jaskelioff, M.; Yuan, L.; Ding, Z.; Protopopov, A.; Kost-Alimova, M.; Hu, J. Utilizing Murine Inducible Telomerase Alleles in the Studies of Tissue Degeneration/Regeneration and Cancer. *J. Vis. Exp. JoVE* **2015**. [[CrossRef](#)]
119. Counter, C.M.; Hirte, H.W.; Bacchetti, S.; Harley, C.B. Telomerase Activity in Human Ovarian Carcinoma. *Proc. Natl. Acad. Sci. USA* **1994**, *91*, 2900–2904. [[CrossRef](#)] [[PubMed](#)]
120. Baykal, A.; Thompson, J.A.; Xu, X.-C.; Hahn, W.C.; Deavers, M.T.; Malpica, A.; Gershenson, D.M.; Silva, E.G.; Liu, J. In Situ Human Telomerase Reverse Transcriptase Expression Pattern in Normal and Neoplastic Ovarian Tissues. *Oncol. Rep.* **2004**, *11*, 297–302. [[CrossRef](#)] [[PubMed](#)]
121. Sasaki, R.; Narisawa-Saito, M.; Yugawa, T.; Fujita, M.; Tashiro, H.; Katabuchi, H.; Kiyono, T. Oncogenic Transformation of Human Ovarian Surface Epithelial Cells with Defined Cellular Oncogenes. *Carcinogenesis* **2009**, *30*, 423–431. [[CrossRef](#)]
122. Osborne, C.K.; Schiff, R. Estrogen-Receptor Biology: Continuing Progress and Therapeutic Implications. *J. Clin. Oncol. Off. J. Am. Soc. Clin. Oncol.* **2005**, *23*, 1616–1622. [[CrossRef](#)] [[PubMed](#)]
123. Li, Y.F.; Hu, W.; Fu, S.Q.; Li, J.D.; Liu, J.H.; Kavanagh, J.J. Aromatase Inhibitors in Ovarian Cancer: Is There a Role? *Int. J. Gynecol. Cancer Off. J. Int. Gynecol. Cancer Soc.* **2008**, *18*, 600–614. [[CrossRef](#)] [[PubMed](#)]
124. Li, H.; Simpson, E.R.; Liu, J.-P. Oestrogen, Telomerase, Ovarian Ageing and Cancer. *Clin. Exp. Pharmacol. Physiol.* **2010**, *37*, 78–82. [[CrossRef](#)] [[PubMed](#)]
125. Losi, L.; Botticelli, L.; Garagnani, L.; Fabbiani, L.; Panini, R.; Gallo, G.; Sabbatini, R.; Maiorana, A.; Benhattar, J. TERT Promoter Methylation and Protein Expression as Predictive Biomarkers for Recurrence Risk in Patients with Serous Borderline Ovarian Tumours. *Pathology* **2021**, *53*, 187–192. [[CrossRef](#)] [[PubMed](#)]

126. Ghareghomi, S.; Ahmadian, S.; Zarghami, N.; Hemmati, S. HTERT-Molecular Targeted Therapy of Ovarian Cancer Cells via Folate-Functionalized PLGA Nanoparticles Co-Loaded with MNPs/SiRNA/Wortmannin. *Life Sci.* **2021**, *277*, 119621. [[CrossRef](#)]
127. Devereux, T.R.; Horikawa, I.; Anna, C.H.; Annab, L.A.; Afshari, C.A.; Barrett, J.C. DNA Methylation Analysis of the Promoter Region of the Human Telomerase Reverse Transcriptase (HTERT) Gene. *Cancer Res.* **1999**, *59*, 6087–6090.
128. Boccardi, V.; Paolisso, G.; Mecocci, P. Nutrition and Lifestyle in Healthy Aging: The Telomerase Challenge. *Aging* **2016**, *8*, 12–15. [[CrossRef](#)]
129. Widschwendter, A.; Müller, H.M.; Hubalek, M.M.; Wiedemair, A.; Fiegl, H.; Goebel, G.; Mueller-Holzner, E.; Marth, C.; Widschwendter, M. Methylation Status and Expression of Human Telomerase Reverse Transcriptase in Ovarian and Cervical Cancer. *Gynecol. Oncol.* **2004**, *93*, 407–416. [[CrossRef](#)]
130. Rafie, N.; Golpour Hamedani, S.; Barak, F.; Safavi, S.M.; Miraghajani, M. Dietary Patterns, Food Groups and Telomere Length: A Systematic Review of Current Studies. *Eur. J. Clin. Nutr.* **2017**, *71*, 151–158. [[CrossRef](#)]
131. Davinelli, S.; Trichopoulou, A.; Corbi, G.; De Vivo, I.; Scapagnini, G. The Potential Nutrigenoprotective Role of Mediterranean Diet and Its Functional Components on Telomere Length Dynamics. *Ageing Res. Rev.* **2019**, *49*, 1–10. [[CrossRef](#)]
132. Pepper, G.V.; Bateson, M.; Nettle, D. Telomeres as Integrative Markers of Exposure to Stress and Adversity: A Systematic Review and Meta-Analysis. *R. Soc. Open Sci.* **2018**, *5*, 180744. [[CrossRef](#)]
133. Canudas, S.; Becerra-Tomás, N.; Hernández-Alonso, P.; Galié, S.; Leung, C.; Crous-Bou, M.; De Vivo, I.; Gao, Y.; Gu, Y.; Meinilä, J.; et al. Mediterranean Diet and Telomere Length: A Systematic Review and Meta-Analysis. *Adv. Nutr.* **2020**, *11*, 1544–1554. [[CrossRef](#)]
134. Navarro-Ibarra, M.J.; Hernández, J.; Caire-Juvera, G. Diet, Physical Activity and Telomere Length in Adults. *Nutr. Hosp.* **2019**, *36*, 1403–1417. [[CrossRef](#)] [[PubMed](#)]
135. Sreerangaraja Urs, D.B.; Wu, W.-H.; Komrskova, K.; Postlerova, P.; Lin, Y.-F.; Tzeng, C.-R.; Kao, S.-H. Mitochondrial Function in Modulating Human Granulosa Cell Steroidogenesis and Female Fertility. *Int. J. Mol. Sci.* **2020**, *21*, 3592. [[CrossRef](#)] [[PubMed](#)]
136. May-Panloup, P.; Boucret, L.; Chao de la Barca, J.-M.; Desquiret-Dumas, V.; Ferré-L'Hotellier, V.; Morinière, C.; Descamps, P.; Procaccio, V.; Reynier, P. Ovarian Ageing: The Role of Mitochondria in Oocytes and Follicles. *Hum. Reprod. Update* **2016**, *22*, 725–743. [[CrossRef](#)] [[PubMed](#)]