

Advanced age – a critical risk factor for recurrent miscarriage

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Abstract: Recurrent spontaneous abortion (RSA) is a multifactorial disease that seriously affects women's physical and mental health. With the advent of efficient contraception, the trend for women towards later maternity until their thirties or even forties. Nevertheless, the risk of miscarriage is strongly related to maternal age. We performed a retrospective analysis to evaluate the etiology of RSA through age groups. The results demonstrated that intrauterine adhesions and ovarian dysfunction were responsible for increased miscarriages in older RSA patients. In conclusion, older women will bear a higher risk of miscarriage, mainly due to uterine adhesions or decreased ovarian function.

Keywords: maternal age, recurrent spontaneous abortion, etiology, outcome of pregnancy

Recurrent spontaneous abortion (RSA) is a multifactorial disease that seriously affects women's physical and mental health (1). The average population of women with one pregnancy loss is 10.8%, among which approximately 1.9% have experienced two consecutive miscarriages, and 0.7% have gone through three or more (2). Different national guidelines have different definitions of RSA in terms of the number of abortions, gestational age, and continuity of abortions (2-4). The Chinese expert consensus released in 2022 defined RSA as the loss of two or more failed clinical pregnancies before 28 weeks with the same sexual partner and pointed out that the occurrence of two consecutive abortions should be paid attention to and evaluated (4).

Nowadays, with the advent of efficient contraception, the trend for women towards later maternity until their thirties or even forties (5); however, the risk of miscarriage depends on the defined upper maternal age (6). The risk rises nearly linearly after age 30 to reach 54% at ages 45 and over, along with decreased egg quality and quantity sharply and an increased rate of mitochondrial DNA mutation and meiosis error (6). What's more, the rate of embryo chromosome abnormality in older women is much higher than in women of appropriate age (7). Due to the relatively high incidence rate of miscarriage, advanced maternal age has become one of the significant

challenges for reproductive medicine. Here, we also designed a retrospective study to evaluate the etiology of RSA based on age grouping, which will benefit pregnant women at an advanced age.

The retrospective study includes 387 RSA patients, which were categorized into two groups depending on age: An older group (≥ 35 years older, $n = 83$) and a younger group (< 35 years older, $n = 304$). There was no significant difference in the body mass index (BMI) and the number of miscarriages between the two groups ($p > 0.05$) (Supplemental Table S1, <https://www.globalhealthmedicine.com/site/supplementaldata.html?ID=69>). Our results demonstrated that the older RSA patients had a higher incidence of intrauterine adhesions (IUA) and ovarian dysfunction than the younger counterparts ($p < 0.05$) due to aging and intrauterine surgery (Table 1), consistent with a previous study (8,9). IUA, a common gynecological and reproductive function uterine disease, strongly influences women's health, becoming one of the leading causes of menstrual loss, abortion, and secondary infertility. All IUA patients are high-risk pregnancies, which is associated with uterine deformation, volume reduction, or insufficient endometrial blood supply caused by IUA, easily leading to post-pregnancy abortion (10). Furthermore, the risk of IUA increases with the number of miscarriage procedures performed

Table 1. The detailed comparison of RSA causes in the different age groups

Causes of RSA	Aged < 35 (n = 304)	Aged ≥ 35 (n = 83)	p value
Chromosome (%)	11 (3.6)	4 (4.8)	0.538
Anatomy (%)	10 (3.3)	10 (12)	0.003*
Bicornuate uterus	1 (0.3)	0	1.000
Mediastinal uterus	4 (1.3)	2 (2.4)	0.613
Adenomyosis	0	1 (1.2)	0.214
Uterine fibroids	1 (0.3)	0	1.000
Intrauterine adhesions	3 (1.0)	6 (7.2)	0.004*
Cervical insufficiency	1 (0.3)	1 (1.2)	0.383
Endocrine (%)	71 (23.4)	21 (25.3)	0.771
Hyperthyroidism	5 (1.6)	2 (2.4)	0.646
Hypothyroidism	5 (1.6)	2 (2.4)	0.646
Subclinical hypothyroidism	27 (8.9)	7 (8.4)	0.898
Polycystic ovary syndrome	6 (2.0)	1 (1.2)	1.000
Decreased ovarian function	1 (0.3)	8 (9.6)	0.000*
Impaired glucose tolerance	5 (1.6)	0	0.589
Insulin resistance	34 (11.2)	4 (4.8)	0.097
Autoimmune (%)	74 (24.3)	23 (27.7)	0.568
AsAb	10 (3.3)	1 (1.2)	0.469
EMAb	9 (3.0)	3 (3.6)	0.726
TPOAb	20 (6.6)	5 (6.0)	1.000
TGAb	14 (4.6)	7 (8.4)	0.177
ACA	6 (2.0)	1 (1.2)	1.000
α-β2GP1	27 (8.9)	10 (12.0)	0.401
ANA	11 (3.6)	4 (4.8)	0.538
Coagulation (%)	19 (6.3)	2 (2.4)	0.272
Higher D-dimer	12 (3.9)	1 (1.2)	0.315
Hyperhomocysteinemia	7 (2.3)	1 (1.2)	1.000
Infection (%)	35 (11.5)	5 (6.0)	0.161
Unexplained (%)	146 (48.0)	34 (41.0)	0.266

Data are n (%); RSA: recurrent spontaneous abortion; AsAb: anti-sperm antibody; EMAb: anti-endometrial antibody; TPOAb: anti-thyroid peroxidase antibody; TGAb: anti-thyroglobulin antibody; ACA: anti-cardiolipin antibodies; anti-β2GP1: anti-β2-glycoprotein 1 antibody; ANA: antinuclear antibody; *p < 0.05.

(11). There were no statistically significant differences in chromosomal abnormalities, endocrine abnormalities, autoimmune abnormalities, coagulation abnormalities, and infection factors between the two groups (Table 1). Hormone levels are significant factors affecting the success of embryo implantation and pregnancy rates. As shown in (Supplemental Table S2, <https://www.globalhealthmedicine.com/site/supplementaldata.html?ID=69>), the level of AMH in the older patients with three or more abortions was significantly lower than that in the older sporadic abortion group ($p < 0.05$), in agreement with the previous result that a low AMH level is associated with high abortion rates in women older than 34 years old (12).

This study investigated the etiology composition of RSA based on age factors. In conclusion, intrauterine adhesions and ovarian dysfunction increased the number of abortions in older RSA patients (≥ 35 years older). More research is needed to elucidate the etiology and mechanisms for recurrent spontaneous abortion.

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References

1. Practice Committee of the American Society for Reproductive Medicine. Definitions of infertility and recurrent pregnancy loss: A committee opinion. *Fertil Steril.* 2020; 113:533-535.
2. Quenby S, Gallos ID, Dhillon-Smith RK, *et al.* Miscarriage matters: The epidemiological, physical, psychological, and economic costs of early pregnancy loss. *Lancet.* 2021; 397:1658-1667.
3. ESHRE Guideline Group on RPL, Bender Atik R, Christiansen OB, Elson J, Kolte AM, Lewis S, Middeldorp S, Nelen W, Peramo B, Quenby S, Vermeulen N, Goddijn M. ESHRE guideline: Recurrent pregnancy loss. *Hum Reprod Open.* 2018; 2018:hoy004.
4. Obstetrics Subgroup, Chinese Society of Obstetrics and Gynecology, Chinese Medical Association, Chinese Expert Consensus Group on Diagnosis and Management of Recurrent Spontaneous Abortion. Chinese expert consensus on diagnosis and management of recurrent spontaneous abortion (2022). *Zhonghua Fu Chan Ke Za Zhi.* 2022; 57:653-667. (in Chinese)
5. Bedenk J, Vrtačnik-Bokal E, Virant-Klun I. The role of anti-Müllerian hormone (AMH) in ovarian disease and infertility. *J Assist Reprod Genet.* 2020; 37:89-100.
6. Magnus MC, Wilcox AJ, Morken NH, Weinberg CR, Häberg SE. Role of maternal age and pregnancy history in risk of miscarriage: Prospective register based study. *BMJ.* 2019; 364:l869.
7. Huang Y, Tu M, Qian Y, *et al.* Age-dependent metabolomic profile of the follicular fluids from women undergoing assisted reproductive technology treatment. *Front Endocrinol (Lausanne).* 2022; 13:818888.
8. Bunnewell SJ, Honess ER, Karia AM, Keay SD, Al Wattar BH, Quenby S. Diminished ovarian reserve in recurrent pregnancy loss: A systematic review and meta-analysis. *Fertil Steril.* 2020; 113:818-827.
9. Carbonnel M, Pirtea P, de Ziegler D, Ayoubi JM. Uterine factors in recurrent pregnancy losses. *Fertil Steril.* 2021; 115:538-545.
10. Li B, Zhang Q, Sun J, Lai D. Human amniotic epithelial

- cells improve fertility in an intrauterine adhesion mouse model. *Stem Cell Res Ther.* 2019; 10:257.
11. Hooker AB, de Leeuw RA, Emanuel MH, Mijatovic V, Brolmann HAM, Huirne JAF. The link between intrauterine adhesions and impaired reproductive performance: A systematic review of the literature. *BMC Pregnancy Childbirth.* 2022; 22:837.
 12. Zarek SM, Mitchell EM, Sjaarda LA, Mumford SL, Silver RM, Stanford JB, Galai N, Schliep KC, Radin RG, Plowden TC, DeCherney AH, Schisterman EF. Antimüllerian hormone and pregnancy loss from the effects of aspirin in gestation and reproduction trial. *Fertil Steril.* 2016; 105:946-952.e942.

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