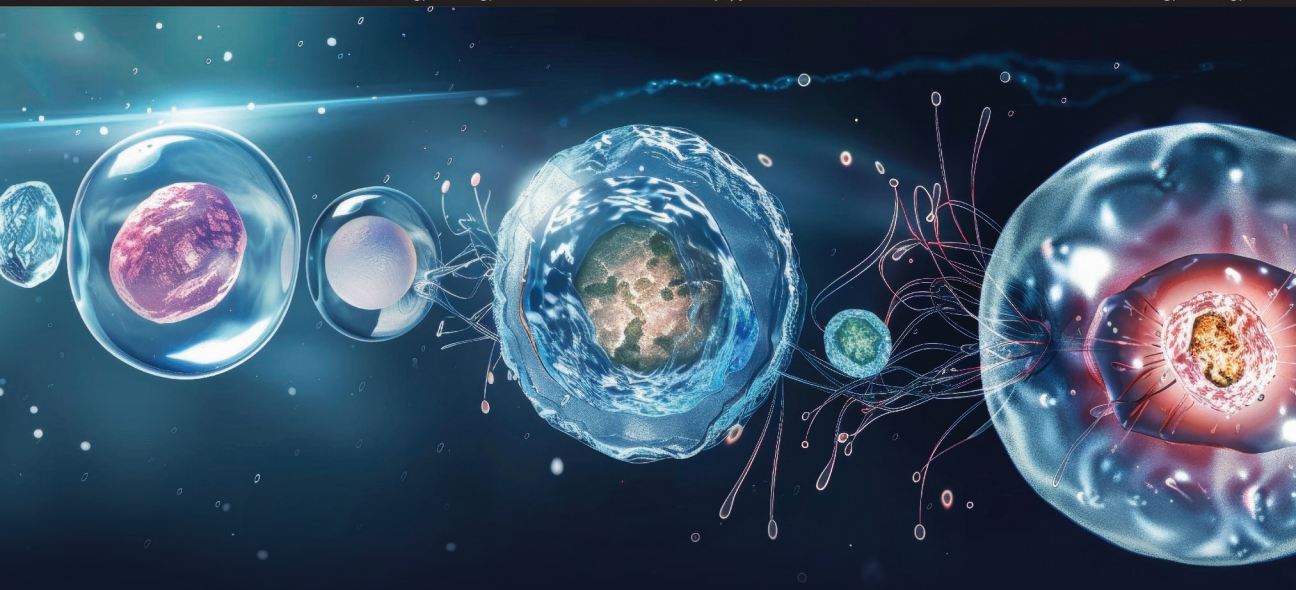
eebrothers.com, for detailed information on obstetrics & gynecology books, visit our website www.jaypeebrothers.com, for detailed information on obstetrics & gynecology books



Troubleshooting in Assisted Reproductive Technology

Kamini Patel

Forewords
Alpesh Gandhi
Sunil Shah
Kanthi Bansal
Jatin P Shah

JAYPEE

Contents

SECTION A: TROUBLESHOOTING IN ASSISTED REPRODUCTIVE TECHNOLOGY: CLINICIAN'S INSIGHTS

1. **Troubleshooting in Patient Selection** 3
   Kamini Patel, Saksha Patel
2. **Troubleshooting in Stimulation in Assisted Reproductive Technology** 24
   Asha Baxi, Dhaval Baxi, Pallavi Pant
3. **Troubleshooting in Ovulation Trigger in Assisted Reproductive Technology** 30
   Kamini Patel, Vani Patel
4. **Troubleshooting in Oocyte Pickup** 46
   Seema Pandey, Karuna Batheja
5. **Troubleshooting in Endometrial Priming** 57
   Nayana Patel, Niket Patel, Molina Patel, Paresh Ghoghari, Harsha Bhadarka, Piyush Chudasama, Kairavi Vyas
6. **Troubleshooting in Embryo Transfer** 74
   Sushma Baxi
7. **Troubleshooting in Recurrent Pregnancy Loss** 81
   Nisarg Patel, Himali Maniar
8. **Troubleshooting in Polycystic Ovary Syndrome** 87
   Himali Maniar, Nisarg Patel

SECTION B: TROUBLESHOOTING IN ASSISTED REPRODUCTIVE TECHNOLOGY: EMBRYOLOGIST'S INSIGHTS

9. **Troubleshooting in Semen Preparation** 97
   Doel Bose Pande, Hemant Deshmukh
10. **Troubleshooting in Conventional In Vitro Fertilization** 110
    D Swaminathan, Pankaj Kaingade

11. Troubleshooting in Laboratory Maintenance and Quality Assurance/Quality Control 116
Viren Patel

12. Troubleshooting in Cryopreservation 127
Harsha Bhadarka

13. Troubleshooting in Intracytoplasmic Sperm Injection 135
Chandan N

14. Troubleshooting in Data Record I 157
Waseem Andrabi, Tabassum Hashmi, Viren Patel

15. Troubleshooter in Data Record II 165
Pankaj Kaingade, Rucha Samant

Index 171

CHAPTER 7

Troubleshooting in Recurrent Pregnancy Loss

Nisarg Patel, Himali Maniar

According to recent studies, the prevalence of recurrent pregnancy loss (RPL) in India is estimated to be around 7.4% based on data from the National Family Health Survey (NFHS), indicating a significant burden of RPL within the country.

"Life affords no greater responsibility, no greater privilege, than the raising of the next generation."
—C Everett Koop

INTRODUCTION

Recurrent pregnancy loss (RPL) poses a challenging scenario in assisted reproductive technology (ART). Although sporadic first-trimester miscarriages remain relatively common, recurrent losses—classically defined as three or more losses before 24 weeks gestation—have unique clinical, psychological, and investigative pathways. This chapter provides a concise yet comprehensive overview of RPL, emphasizing practical troubleshooting tips for busy clinicians. Drawing on major guidelines, including those from the Royal College of Obstetricians and Gynaecologists (RCOG), the American Society for Reproductive Medicine (ASRM), and the European Society of Human Reproduction and Embryology (ESHRE), this chapter will discuss how best to investigate and manage women and couples affected by RPL in the ART setting.[1-4]

EPIDEMIOLOGY AND DEFINITIONS

- *Traditional RPL definition:* Three or more consecutive first-trimester losses or one or more second-trimester miscarriages. However, many clinicians consider early evaluation after two losses if the suspicion of a pathological cause is high.[5-8]
- *Incidence:* About 1–3% of couples experience RPL, although the true rate can vary with population characteristics and evolving definitions.
- *Shift in perspective:* There is growing recognition that even two miscarriages—particularly if they are confirmed euploid or clinically significant—may warrant thorough evaluation.[9,10]

CLINICAL SIGNIFICANCE AND IMPACT

- RPL results in considerable emotional distress, necessitating empathetic

JAYPEE BROTHERS

counseling and a comprehensive management plan.
- Even in dedicated recurrent miscarriage clinics, over half of couples may remain with an "unexplained" diagnosis, making supportive care and thorough investigation key components of care.

In the following sections, we outline a practical, stepwise approach to RPL. We begin with key areas of troubleshooting, including epidemiological and maternal risk factors, uterine anomalies, endocrine disorders, and immunological aspects, then move to management strategies.

COMMON RISK FACTORS AND INITIAL TROUBLESHOOTING (TABLE 1)

Table 1 summarizes some major established risk factors for RPL. Clinicians should consider each factor during history-taking and initial workup.

Practical Tips

- Ensure modifiable risk factors (e.g., weight, smoking, and caffeine) are addressed early.
- Offer sensitive counseling when advanced maternal age or paternal age is identified.
- Investigate broader lifestyle and environmental exposures if RPL remains otherwise unexplained.

TABLE 1: Major established risk factors for RPL.

Risk factor	*Association/keynotes*
Advancing maternal age	Strong link; higher aneuploidy rates beyond the age of 35 years
Paternal age >40	Mild to moderate effect; less pronounced than maternal age
Obesity (BMI >25 kg/m^2)	Raises odds of miscarriage and obstetric complications
Chronic smoking/ excess alcohol	Linked to higher miscarriage incidence
Excess caffeine (>200–300 mg/ day)	Observational data suggest increased risk, though some controversy remains.
Consanguinity	Uncertain link; conflicting small-scale data
Black ethnic background	Some observational studies show higher odds of sporadic miscarriage
Number of previous miscarriages	Each additional prior loss raises the risk of a subsequent miscarriage

(BMI: body mass index; RPL: recurrent pregnancy loss)
Source: Adapted from references[1-4].

ANATOMICAL CONSIDERATIONS

Uterine Anomalies

Structural uterine anomalies—congenital (e.g., septate and bicornuate) or acquired (e.g., submucosal fibroids and intrauterine adhesions)—carry the potential to reduce endometrial receptivity or increase midtrimester losses.

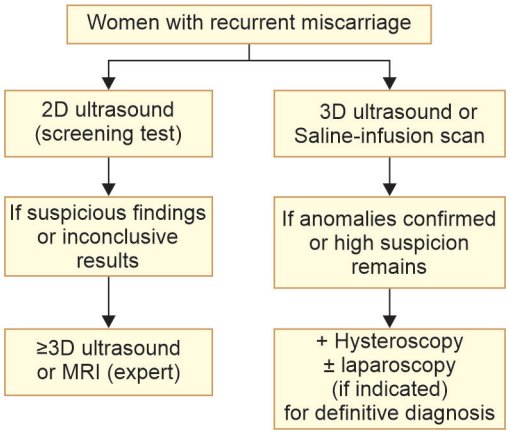
- *Congenital uterine anomalies:* Septate and bicornuate uteri are most frequently implicated.
  - *Septate uterus:* Often associated with poor pregnancy outcomes in repeated first or second-trimester losses.
  - *Bicornuate uterus:* Linked to second-trimester losses, although data vary.
- *Acquired uterine anomalies:* Submucosal fibroids, polyps, or Asherman's syndrome (intrauterine adhesions) may contribute to RPL.

Flowchart 1 outlines a recommended imaging pathway.

- *Troubleshooting tip:* 3D ultrasound is the gold standard noninvasive investigation

JAYPEE BROTHERS

Flowchart 1: Imaging approach for suspected uterine anomalies.



for congenital anomalies; MRI helps clarify complex cases.

- *Clinical pearl:* Resection of a significant septum can improve live birth rates, ideally performed within an appropriate evidence-based framework.

GENETIC FACTORS AND CHROMOSOMAL TESTING

- *Parental chromosomal rearrangements:* Approximately 2–5% of couples with RPL have a translocation or inversion. Routine parental karyotyping after three losses is standard, but some centers test after two.
- *Fetal chromosomal anomalies:* Chromosomal errors in miscarriage tissue remain the most common cause of early pregnancy loss. Up to ~40–50% of sporadic losses have aneuploidy, though the rate in RPL may be slightly lower (~30–40%).
- *Troubleshooting:*
  - Test pregnancy tissue (e.g., via microarray) whenever possible.
  - If an unbalanced translocation is found in miscarriage tissue, check parental karyotypes to identify carriers.
  - Offer genetic counseling to translocation carriers regarding reproductive choices, including the possibility of preimplantation genetic testing for structural rearrangements (PGT-SR) or donor gametes.

ENDOCRINE AND METABOLIC DISORDERS

Thyroid Dysfunction

- Overt hypothyroidism is strongly linked to miscarriage risk. Treat with levothyroxine to maintain thyroid-stimulating hormone (TSH) in the lower-normal range.
- *Subclinical hypothyroidism (SCH)* (TSH >4 mIU/L especially) may carry increased miscarriage risk, though controversies remain regarding TSH thresholds (2.5 vs. 4.0 mIU/L).
- *Thyroid peroxidase (TPO)*-antibodies in euthyroid women appear associated with higher RPL incidence, but robust evidence for routine thyroxine therapy in all euthyroid, TPO-positive women is lacking.

Polycystic Ovary Syndrome

Women with polycystic ovary syndrome (PCOS) often exhibit hyperinsulinemia and hyperandrogenemia that can predispose to poor pregnancy outcomes. However, direct evidence linking PCOS to RPL remains somewhat inconsistent. Clinicians may consider metformin in those with insulin resistance or elevated free androgen index.

Other Considerations

- *Obesity:* Weight reduction can significantly improve fertility and reduce miscarriage risk.
- *Diabetes:* Optimize glycemic control to lower miscarriage rates and congenital anomalies.

JAYPEE BROTHERS

- *Prolactin imbalances:* Mild hyperprolactinemia may interfere with corpus luteum function.

Bromocriptine or cabergoline could be beneficial if significantly elevated prolactin is confirmed.

Key Troubleshooting Pearl

Reevaluate borderline TSH or borderline elevated prolactin in repeated tests, ensuring no laboratory or timing artifacts.

IMMUNOLOGICAL AND THROMBOPHILIC FACTORS

Acquired Thrombophilia: Antiphospholipid Syndrome

- Defined by RPL plus persistently positive antiphospholipid (aPL) antibodies (lupus anticoagulant or anticardiolipin antibodies).
- *Aspirin + heparin* [either unfractionated or low-molecular-weight heparin (LMWH)] is the only evidence-based therapy that significantly improves live birth rates in antibody syndrome (APS).
- *Practical caveat:* APS remains a leading cause of *treatable* RPL; confirm at least 12 weeks apart for consistent positivity.

Inherited Thrombophilia

- Includes Factor V Leiden, prothrombin gene mutation, and deficiencies in protein C/S or antithrombin.
- The link with RPL is weaker compared to APS, particularly for first-trimester losses; prophylactic LMWH or aspirin shows no uniform benefit unless there are additional risk factors (e.g., personal VTE history).

Troubleshooting

- Discuss testing limitations with couples; false positives and questionable associations can complicate management.
- If identified, weigh maternal risk (thrombosis) and pregnancy context before prescribing prophylaxis.

UNEXPLAINED RECURRENT PREGNANCY LOSS AND ADDITIONAL MANAGEMENT CONSIDERATIONS

Preimplantation Genetic Testing for Aneuploidy or In Vitro Fertilization for Unexplained Recurrent Pregnancy Loss

- Preimplantation genetic testing for aneuploidy (PGT-A) has not consistently shown an improved live birth rate among couples without known chromosomal rearrangements; the cost and procedure burden may outweigh the benefits.
- Assisted conception is not automatically more successful if RPL is unexplained unless other infertility factors exist.

Progesterone Therapy

- Large trials (PROMISE and PRISM) indicate that universal first-trimester progesterone supplementation does not strongly impact RPL outcome. However, in women presenting with bleeding in early pregnancy plus a history of recurrent losses, vaginal micronized progesterone may offer a modest benefit in live birth rates.
- *Practical pearl:* Administer 400 mg micronized progesterone twice daily from bleeding onset until 16 weeks if indicated.

JAYPEE BROTHERS

Supportive Care and Psychological Considerations

- Dedicated RPL clinics offering reassurance scans and consistent physician follow-up have been linked to increased satisfaction and potentially improved outcomes.
- Emotional distress, anxiety, and depression rates are high in RPL; timely psychological support is essential.
- Encourage couples to consider counseling or therapy to manage grief and anxiety, especially after repeated losses.

PUTTING IT ALL TOGETHER: A STEPWISE MANAGEMENT (BOX 1)

BOX 1: Stepwise management of recurrent miscarriage.

- *Confirm RPL definition:*
  - ≥3 consecutive first-trimester losses, or
  - ≥1 second-trimester loss
- *Comprehensive evaluation:*
  - Detailed history and exam (lifestyle, genetics, and medical history)
  - *Ultrasound:* 3D for congenital anomalies
  - *Laboratories:*
    - *APS:* Lupus anticoagulant and aCL
    - Inherited thrombophilia (Factor V Leiden, etc., especially if second trimester)
    - Thyroid function (TSH and TPO)
    - ± Prolactin if suspicion
    - Karyotype pregnancy tissue if possible
  - Parental karyotypes if indicated
- *Management interventions:*
  - *Treat APS:* Aspirin + LMWH
  - *TSH or overt thyroid disease:* Levothyroxine
  - *PCOS:* Metformin if IR or hyperandrogenemia
  - Correct modifiable risk factors (BMI, smoking, EtOH, and caffeine)
  - Consider hysteroscopic correction for the septate uterus
  - Psychological support, repeated scans

Contd...

- *Future pregnancy:*
  - Early contact with RPL team or dedicated clinic
  - ± Progesterone if threatened miscarriage
  - Reassurance scans at 6–7 weeks, then q2–3 weeks if feasible
- *If additional loss occurs:*
  - Reassess potential overlooked factors
  - Check the feasibility of pregnancy tissue testing
  - Continue emotional and psychosocial support

(aCL: anticardiolipin antibody; APS: acquired thrombophilia; BMI: body mass index; ETOH: ethyl alcohol or ethanol; LMWH: low-molecular-weight heparin; RPL: recurrent pregnancy loss; TPO: thyroid peroxidase; TSH: thyroid-stimulating hormone)

CONCLUSION

Recurrent pregnancy loss demands meticulous diagnostics and a tailored approach. While a significant proportion may remain "unexplained," many couples benefit from systematic investigations and interventions that target modifiable risk factors, thrombophilias (especially APS), uterine anomalies, and endocrine imbalances. Crucially, management extends beyond medications and procedures—clinicians should prioritize psychological support and continuity of care. Early pregnancy monitoring, emotional counseling, and robust patient education can enhance both clinical success rates and patient well-being.

Despite evolving definitions and some debate (e.g., the threshold at which to initiate investigations), consensus remains on the need for thorough evaluation. Adequate preconception and early pregnancy care, specialized imaging, judicious testing for genetic or immunological causes, and empathetic, evidence-based intervention can offer significant hope to couples struggling with repeated pregnancy loss.

JAYPEE BROTHERS

Key Points

- RPL demands meticulous diagnostics and a tailored approach. While a significant proportion may remain "unexplained," many couples benefit from systematic investigations and interventions that target modifiable risk factors, thrombophilias (especially APS), uterine anomalies, and endocrine imbalances.
- Crucially, management extends beyond medications and procedures—clinicians should prioritize psychological support and continuity of care.
- Early pregnancy monitoring, emotional counseling, and robust patient education can enhance both clinical success rates and patient well-being.
- Despite evolving definitions and some debate (e.g., the threshold at which to initiate investigations), the consensus remains on the need for a thorough evaluation.
- Adequate preconception and early pregnancy care, specialized imaging, judicious testing for genetic or immunological causes, and empathetic, evidence-based intervention can offer significant hope to couples struggling with repeated pregnancy loss.

REFERENCES

1. Practice Committee of the American Society for Reproductive Medicine. Evaluation and treatment of recurrent pregnancy loss: a committee opinion. Fertil Steril. 2012;98(5):1103-11.
2. Royal College of Obstetricians and Gynaecologists. (2011). Recurrent and late miscarriage (green-top guideline no. 17). [Online] Available from https://www.rcog.org.uk/guidance/browse-all-guidance/green-top-guidelines/recurrent-miscarriage-green-top-guideline-no-17/ [Last accessed January, 2025].
3. European Society of Human Reproduction and Embryology (ESHRE). (2017). Guideline on the management of recurrent pregnancy loss. [Online] Available from https://www.eshre.eu/Guidelines-and-Legal/Guidelines/Recurrent-pregnancy-loss [Last accessed January, 2025].
4. Quenby S, Gallos ID, Dhillon-Smith RK, Podesek M, Stephenson MD, Fisher J, et al. Miscarriage matters: the epidemiological, physical, psychological, and economic costs of early pregnancy loss. Lancet. 2021;397(10285):1658-67.
5. Saravelos SH, Li TC. Unexplained recurrent miscarriage: how can we explain it? Hum Reprod. 2012;27(7):1882-6.
6. Coomarasamy A, Devall AJ, Brosens JJ, Quenby S, Stephenson MD, Sierra S, et al. Micronized vaginal progesterone to prevent miscarriage: a critical evaluation of randomized evidence. Am J Obstet Gynecol. 2020;223(2):167-76.
7. Empson M, Lassere M, Craig J, Scott J. Prevention of recurrent miscarriage for women with antiphospholipid antibody or lupus anticoagulant. Cochrane Database Syst Rev. 2005;2005(2):CD002859.
8. Smits MA, van Maarle M, Hamer G, Mastenbroek S, Goddijn M, van Wely M. Cytogenetic testing of pregnancy loss tissue: a meta-analysis. Reprod Biomed Online. 2020;40(6):867-79.
9. Coomarasamy A, Dhillon-Smith RK, Papadopoulou A, Al-Memar M, Brewin J, Abrahams VM, et al. Recurrent miscarriage: evidence to accelerate action. Lancet. 2021;397(10285):1675-82.
10. Li TC, Marren A. Recurrent pregnancy loss: a summary of international evidence-based guidelines and practice. Reprod Biomed Online. 2022;45(7):63-78.

JAYPEE BROTHERS

for detailed information on obstetrics & gynecology books, visit our website www.jaypeebrothers.com, for detailed information on obstetrics & gynecology books, visit our website

Troubleshooting in Assisted Reproductive Technology

ART has revolutionized with greater advancements offering hope to millions of individuals suffering from infertility, even with such revolution in technology and clinical expertise, ART procedures are still quite complex and unpredictable with several variables affecting the success rate. As every individual has a unique fingerprint and every lock has its own key, likewise, the ART cycle for each individual is different. The ART cycle in its own way presents a unique set of challenges, starting from tracking down an individual's menstrual cycle to designing an ovarian stimulation, performing IVF/ICSI to embryo culture, and ultimately navigating embryo transfer. These days not only technology advancements, but the trend of "personalized treatment" is also helping the clinicians and embryologists a lot. However, the success rates are still quite low. Even experienced clinicians and embryologists can encounter unexpected hinderances and failures, alarming the immediate need for effective troubleshooting strategies.

The book *Troubleshooting in Assisted Reproductive Technology* is a comprehensive guide to addressing challenges associated with patient selection, ovarian stimulation, ovulation trigger, ovarian pickup, endometrial priming, embryo transfer, and cases of recurrent pregnancy loss and polycystic ovary syndrome (PCOS) in *Section A: Clinician's Insights,* while *Section B: Embryologist's Insights* addresses challenges associated with semen preparation, conventional IVF, laboratory maintenance-associated IVF outcomes, cryopreservation, ICSI, and maintenance of data record in IVF laboratories.

This book will be of great help to fellow aspiring clinicians who plan to become an IVF specialist and those who face recurrent failures in their course of treatment and clinical embryologists. In addition, it will provide insights into the troubleshooting strategies in the course of treatment for the clinicians and strategies for embryologists to combat-varied hurdles coming in the way of providing top-grade embryos.

After this, we aspire to launch our next edition on how to get good results with multiple IVF failures.



Kamini Patel MBBS DGO DICOG is a highly accomplished an Infertility Specialist and the visionary Founder and Director of Vani IVF Centre, Ahmedabad, Gujarat, India, a premier institution in fertility care with over three decades of excellence. With an impressive 31 years of experience in the field, she has established herself as a trusted name in reproductive health and assisted reproductive technology (ART). Under her leadership, Vani IVF Centre has expanded to five fully operational centers across Gujarat, including three in Ahmedabad, providing advanced fertility treatments and setting benchmarks in patient care. Dr Patel's expertise in addressing cases of recurrent pregnancy loss and recurrent implantation failure has earned her a reputation for achieving some of the highest success rates in these challenging areas. Her contributions to the medical field extend beyond her clinical practice. She served as the President of the Ahmedabad Obstetrics and Gynaecology Society (AOGS) and the Society of Gynaecological Oncology of Gujarat (SOGOG) during 2021–2022, showcasing her leadership and dedication to advancing women's health. In recognition of her exceptional work, she was recently honored with the "Most Promising IVF Chain in Gujarat" award by Times Network.

Additionally, Dr Kamini Patel's expertise was acknowledged at the highest level when she was invited as a panelist to discuss Gujarat's Medical Infrastructure 2025, showcasing her pivotal role in shaping the region's healthcare landscape. She continues to inspire both her peers and patients with her unwavering commitment to excellence, innovation, and compassionate care.

Printed in India

Available at all medical bookstores or buy online at www.ejaypee.com

JAYPEE BROTHERS
Medical Publishers (P) Ltd.
EMCA House, 23/23-B, Ansari Road,
Daryaganj, New Delhi - 110 002, INDIA
www.jaypeebrothers.com

Join us on facebook.com/JaypeeMedicalPublishers
Follow us on instagram.com/JaypeeMedicalPublishers

Shelving Recommendation
OBSTETRICS & GYNECOLOGY

ISBN 978-93-6616-185-3