Mini Review

N-acetyl cysteine in clomiphene citrate resistant polycystic ovary syndrome: A review of reported outcomes

Lekha Saha, Sharonjeet Kaur, Pradip Kumar Saha[1]

Departments of Pharmacology, [1]Obstetrics and Gynaecology, Postgraduate Institute of Medical Education and Research, Chandigarh, India

ABSTRACT

Clomiphene citrate (CC) has been the gold-standard drug for ovulation induction in polycystic ovary syndrome (PCOS), but still CC resistance is seen in approximately 15-40% in women with PCOS. *N*-acetyl cysteine (NAC), a safe and cheap drug available in the market many years ago as mucolytic agent, was found to have a role in infertility management. Recently, some reports discussed the possible beneficial effects of NAC on ovulation. The biological properties of the NAC make this drug a potential candidate for its use in the infertility treatment, especially in the PCOS in inducing or augmenting ovulation. An updated electronic search was performed through PUBMED, MEDLINE, and COCHRANE and focused on peer-reviewed, full text, randomized controlled trials, and observational cohort or case-control studies for role of NAC in CC-resistant PCOS. Thorough search through all the clinical studies showed mixed results. Studies with positive results showed improvement in induction of ovulation as compared to negative studies showing contrary results. More randomized clinical trials are still needed to establish its definitive role in CC-resistant PCOS.

Key words: Clomiphene citrate resistant, *N*-acetyl cysteine, polycystic ovary syndrome, review

INTRODUCTION

The first description of the polycystic ovary syndrome (PCOS) dates back to the original report of seven cases by Stein and Leventhal in 1935.[1] PCOS is now widely accepted as a syndrome, a constellation of clinically recognizable features, signs, and symptoms. However, there remains a lack of consensus about how to define and diagnose this syndrome. In the last two decades, three alternative definitions have been formulated for the diagnosis of PCOS. The most widely used 1990 National Institutes of Health (NIH) criteria include clinical and/or biochemical hyperandrogenism and chronic anovulation.[1] The 2004 Rotterdam criteria suggest PCOS should be diagnosed by two of the following three criteria: oligo-anovulation, clinical or biochemical hyperandrogenism, and polycystic ovaries on ultrasound.[1] The most recent androgen excess and PCOS (AE–PCOS) Society criteria recommend that PCOS should be defined as clinical or biochemical hyperandrogenism associated with ovulatory dysfunction in the form of oligo-anovulation or polycystic ovaries on ultrasound.[1] All three sets of criteria highlight exclusion of other related disorders before making a diagnosis of PCOS.

Access this article online	
Quick Response Code:	Website: www.jpharmacol.com
	DOI: 10.4103/0976-500X.114597

Previous studies assess the prevalence of PCOS in different populations with reported rates between 4% and 8% using

Address for correspondence:
Lekha Saha, Department of Pharmacology, Postgraduate Institute of Medical Education and Research, Chandigarh - 160 012, India. E-mail: lekhasaha@rediffmail.com

NIH criteria, between 2.4% and 11.9% using Rotterdam criteria, and between 2.2% and 10.2% using AE–PCOS Society criteria. Variation in the rates among the studies using the same diagnostic criteria might be due to differences in background study populations, difficulties in phenotypic definition, and design limitations including biased sampling. More recent study conducted by Yildiz *et al*. concluded that the prevalence of PCOS under NIH, Rotterdam, and AE–PCOS Society criteria are 6.1%, 19.9%, and 15.3%, respectively.[1]

Clomiphene citrate (CC) remains the treatment of first choice for induction of ovulation in anovulatory women with PCOS. Cost of medication is low, the oral route of administration is patient friendly, there are relatively few adverse effects, little ovarian response monitoring is required, and abundant clinical data are available regarding safety of the drug.[2] The mechanism of action is not entirely known, but it is thought to involve the blockade of the negative feedback mechanism that results in increased secretion of follicle-stimulating hormone (FSH).[2] The main factors that predict outcome of treatment are obesity, hyperandrogenemia, and age. Ovarian volume and menstrual status are additional factors that help to predict responsiveness to CC.[2]

The starting dose of CC generally should be 50 mg/day (for 5 days, starting on day 2-5, following a spontaneous or progestin-induced withdrawal bleeding) and can be increased to 250 mg/day.[2] The doses of CC in excess of 100 mg/day are not approved by the US FDA.[3] However, managed care studies have shown that the most effective dosage is 100-150 mg/day and over 75% of ovulations occur within these dosages.[2] After six to nine cycles of treatment with CC, cumulative pregnancy rates reach 70-75%.[2] Life table analysis of the most reliable studies indicated a conception rate up to 22% per cycle in women ovulating on CC.[2] Treatment generally should be limited to six (ovulatory) cycles.[2,4]

Clomiphene resistance defined as failure to ovulate after receiving 150 mg of CC daily for 5 days per cycle, for at least three cycles, is common and occurs in approximately 15-40% in women with PCOS.[5] Insulin resistance, hyperandrogenemia, and obesity represent the major factors involved in CC resistance; avert the ovaries from responding to raised endogenous FSH levels following CC therapy.[6] Moreover, a genetic predisposition was suggested.[7]

Insulin resistance with compensatory hyperinsulinemia is a prominent feature of the syndrome and appears to have a patho-physiological role in the hyperandrogenism of the disorder. Hyperinsulinemia results in increased ovarian androgen biosynthesis and decreased sex-hormone-binding globulin synthesis from the liver, leading to increased bioavailability of free androgen. The excess in local ovarian androgen augmented by hyperinsulinemia causes premature follicular atresia and anovulation.[6]

N-acetyl cysteine

N-acetyl cysteine (NAC) is the acetylated variant of the amino acid L-cysteine. It is an excellent source of sulfhydryl groups and is converted *in vivo* into metabolites that stimulate glutathione production, promote detoxification, and act directly as free-radical scavengers. Historically, NAC is a mucolytic in a variety of respiratory illnesses; however, it appears to be beneficial in other conditions, such as HIV infection, cancers, heart diseases, smoking, heavy-metal poisoning, and prevention of influenza, epilepsy, and acetaminophen poisoning. NAC has different actions inside the body. It is primarily a powerful antioxidant; it has activity on insulin secretion in pancreatic cells and on insulin receptors on human erythrocytes. NAC has antiapoptotic effects; it can preserve vascular integrity and has immunological functions.[8]

Reports in the literature have discussed the possible beneficial effects of NAC on ovulation in CC-resistant PCOS patients.[9,10] Rizk *et al*. have noted that the combination of CC and NAC increases ovulation and pregnancy rates in CC-resistant PCOS patients who also suffer from infertility.[9] In 2007, Badawy *et al*. noted that the addition of NAC to a CC regimen in patients with PCOS would increase ovulation rates significantly.[10] Biological activities of NAC have been depicted in Figure 1. Many mechanisms are proposed to explain the unique action of NAC on ovulation. Borgstrom *et al*. demonstrated the insulin-sensitizing activity of NAC.[11] Flughesu *et al*. found that NAC caused significant reduction in circulating insulin levels, peripheral IR, serum testosterone levels, and free androgen index in PCOS.[12] Other important mechanisms explaining the beneficial effects of NAC rather than its insulin-sensitizing effects were also reported. Odetti *et al*. reported that NAC has antiapoptotic effects on the ovary and apoptosis is definitely responsible for the process of follicular atresia.[13] Youssef *et al.* (2006) evaluated the possible beneficial mucolytic effect of NAC on CC-resistant patients with poor postcoital tests and reported that effect of NAC as an adjuvant to CC appears not to be related to its mucolytic effect.[14]

With the above-mentioned background, the aim of this review is to discuss the available evidences either supporting or against the usage of the NAC in CC-resistant PCOS patients and to make a meaningful conclusion.

Literature search

A search was conducted using PUBMED, MEDLINE, and COCHRANE database and focused on peer-reviewed, full text, randomized controlled trials, and observational cohort or case control studies using the keywords: "NAC," "ovulation," "PCOS," "Clomiphene resistance," and "antioxidant." We

updated our electronic search and the last date of the search updating was at December 2011. We mainly focused on the effect of NAC in PCOS. We excluded studies that were experimental or laboratory based [Figure 2]. We also scanned all references from identified articles for additional relevant information. After all the data had been extracted, a table was constructed with respect to first author, study design, patient settings, study groups, outcome, and conclusion. There was no time or language limit in our literature search.

Evidence supporting the benefit of the NAC in clomiphene-resistant PCOS

Only one randomized trial has been conducted which demonstrated the beneficial role of NAC in clomiphene-resistance PCOS patients [Table 1].

A placebo-controlled double-blind randomized trial was conducted by Rizk *et al.* (2005) and showed that the combination of NAC (1.2 g/d) with CC (100 mg/d) for 5 days significantly increased both ovulation and pregnancy rates in 150 obese women with CC-resistant PCOS (49.3% vs. 1.3% and 21.3% vs. 0, respectively, $P < 0.0001$, $P = 0.00006$).[9]

There are other two studies of Salehpour *et al.* (2009) and Badawy *et al.* (2007) that was done in PCOS patients without CC resistance. Results of a cross-over trial conducted by Badawy *et al.* (2007) showed significant improvement in the ovulation rate, serum oestrogen, progesterone, endometrial thickness, and pregnancy rate when NAC was combined with CC as compared to CC alone in 573 women with PCOS.[10] Study by Salehpour *et al.* (2009) reported improvement in the ovulation rate with NAC (55.60%,) as compared to placebo group (16.7%,) in total of 46 women with PCOS ($P = 0.035$),[15] and there was also a significant reduction in weight and body mass index as well as waist/hip ratio. Various other parameters like fasting blood sugar, serum insulin, total cholesterol, and low-density lipoprotein and Homeostatic model of insulin resistance were decreased and high-density lipoprotein was elevated significantly. No significant improvement in the hormonal profile was observed which included luteinizing hormone, FSH, prolactin, leutinizing hormone/follicle-stimulating hormone (LH/FSH) levels, and glucose/insulin ratio.[15]

It can be concluded from the results of these studies that NAC promotes lipid profile, hormonal levels, ovulation, and consequently, the long-term health status of women with both PCOS and CC-resistant PCOS through inhibition of oxidative stress and improvement of peripheral insulin.

Evidence against the benefit of the NAC in clomiphene-resistant PCOS

Like studies which have demonstrated the beneficial role of NAC in clomiphene resistance PCOS, there are studies which have failed to demonstrate the beneficial effect of NAC in clomiphene resistance PCOS [Table 2]. A prospective randomized controlled study was conducted by Elnashar *et al.* (2007) in 61 infertile women with CC-resistant PCOS. Women were assigned randomly to receive either metformin (1,500 mg/d) or NAC (1.8 g/d) for 6 weeks. In the metformin group, there was a significant decrease in the fasting glucose, fasting insulin, and total testosterone. In the NAC group, there was no significant difference in the fasting glucose or fasting insulin and there was a significant

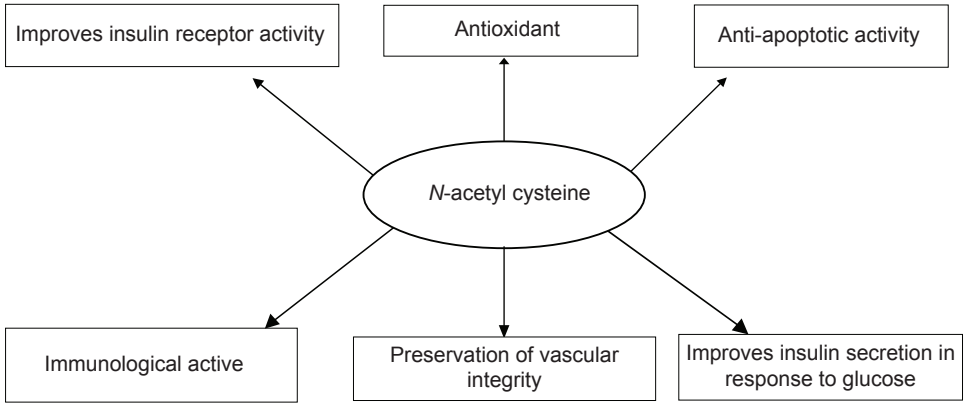

Figure 1: Biological activities of *N*-acetyl cysteine

Table 1: Clinical evidence of use of *N*-acetyl cysteine in clomiphene citrate resistant polycystic ovary syndrome

Study	Study design	*N*	Study group	Primary outcome	Summary of key findings
Rizk *et al.* (2005)[9]	Placebo-controlled double-blind randomized trial	150	1st: NAC+CC 2nd: CC+Placebo	Ovulation Rate	Significant higher ovulation rate (49.3% in NAC as compared to 1.3% in Placebo group)

NAC=*N*-acetyl cysteine, CC=Clomiphene citrate

Table 2: Clinical evidence against the use of *N*-acetyl cysteine in clomiphene citrate resistant polycystic ovary syndrome

Study	Study design	*N*	Study group	Primary outcome	Summary of key findings
Elanashar *et al.* (2007)[16]	Prospective randomized controlled study	61	1st: CC 2nd: NAC	Ovulation rate, plasma insulin levels, and plasma testosterone	Metformin showed significant improvement in ovulation rate (51.6%) as compared to NAC (6.7%)
Youssef *et al.* (2006)[14] Abu Hashim *et al.* (2010)[17]	Prospective controlled pilot study A randomized controlled trial	39 192	NAC+CC 1st NAC+CC 2nd metformin+CC	Clinical pregnancy rate Ovulation	No demonstrable difference in the clinical pregnancy rate between two groups The efficacy of metformin-CC combination therapy is higher than that of NAC-CC for inducing ovulation and achieving pregnancy among CC-resistant PCOS patients

NAC=*N*-acetyl cysteine, CC=Clomiphene citrate

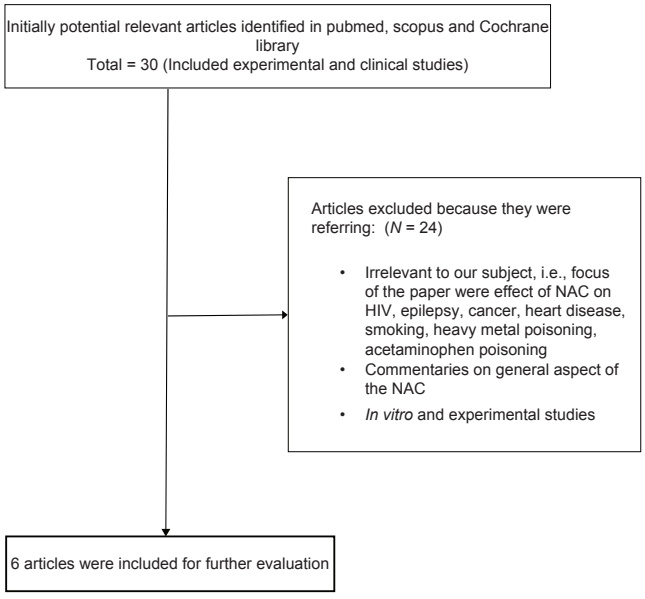

Figure 2: Flow chart of the reviewed articles

decrease in total testosterone. There was no significant difference in the fasting glucose–fasting insulin ratio in both groups. In the metformin group, the rate of ovulation was 51.6% (16/31) vs. 6.7% (2/30) in the NAC group, which was statistically significant.[16] The authors concluded that metformin alone is an effective drug in inducing ovulation in CC-resistant PCOS, whereas NAC alone is not.[16]

Study by Youssef *et al.* (2006) in 39 clomiphene-resistant PCOS was a prospective controlled pilot study and there was no statistical difference between the two groups pertaining the clinical pregnancy rate ($P = 0.24$). The authors acknowledged that the sample size of the study was not large enough to withdraw firm conclusions.[14]

A recent randomized controlled trial by Abu Hashim *et al.* (2010) showed that the efficacy of metformin-CC combination therapy is higher than that of NAC-CC for inducing ovulation and achieving pregnancy among CC-resistant PCOS patients.[17] In this study, 192 women were randomized to receive either the NAC-CC drug combination (NAC-CC, $n = 95$) or the combined metformin-CC therapy (metformin-CC, $n = 97$) for three treatment cycles. The primary outcome measure was ovulation; secondary outcomes were number of follicles, serum estrogen and progesterone levels, post-treatment endometrial thickness, pregnancy, and miscarriage. Over a 3-month follow-up period, women in metformin-CC group had significantly higher ovulation and pregnancy rates compared with women in NAC-CC group (69.1% vs. 20.0%, $P = 0.002$, and 22.7% vs. 5.3%, $P = 0.020$, respectively). The level of serum estrogen, the endometrial thickness on the day of human chorionic gonadotropin administration, and the serum progesterone level on cycle days 21-23 were all significantly higher for women in metformin-CC group than for those in NAC-CC group. Additionally, a lower miscarriage rate was observed among women in metformin-CC group than among those in NAC-CC group.[17]

To summarize, the level of evidences supporting the use of NAC in CC-resistant PCOS patients is poor as compared with that against its usage in them. There is only one randomized controlled trial conducted by Rizk *et al.* (2005) in 150 obese women with CC-resistant PCOS, which showed that the combination of NAC (1.2 g/day) with CC (100 mg/day) for 5 days significantly increased both ovulation and pregnancy rates (49.3% vs. 1.3% and 21.3% vs. 0, respectively, $P < 0.0001$, $P = 0.00006$).[9] Other two studies by Salehpour *et al.* (2009) and Badawy *et al.* (2007) have shown significant improvement in the ovulation rate with NAC but were done in PCOS patients without CC resistance. Evidence against the use of NAC in CC-resistant PCOS women is strong including three randomized controlled trials performed on 292 CC-resistant PCOS patients in total ($192 + 61 + 39 = 292$)[14,16,17] as compared to 150 patients in the study by Rizk *et al.*[9] Dosages and duration of NAC varied between all the studies ranging from 1.2 to 1.8 g/day for 5 days to 5-6 weeks.

CONCLUSION

NAC is an antioxidant, which acts by stimulating the synthesis of glutathione under conditions of excessive oxidative stress or

during certain disease processes. However, studies regarding its utility in PCOS have shown inconsistent results; further randomized clinical trials with large number of CC-resistant PCOS women are still needed to shed more light on this topic.

REFERENCES

1. Yildiz BO, Bozdag G, Yapici Z, Esinler I, Yarali H. Prevalence, phenotype and cardiometabolic risk of polycystic ovary syndrome under different diagnostic criteria. Hum Reprod 2012;27:3067-73.
2. Thessaloniki ESHRE/ASRM-Sponsored PCOS Consensus Workshop Group. Consensus on infertility treatment related to polycystic ovary syndrome. Hum Reprod 2008;23:462-77.
3. Practice Committee of the American Society for Reproductive Medicine. Use of clomiphene citrate in women. Fertil Steril 2006;86(Suppl 4):S187-93.
4. Homburg R. Clomiphene citrate-end of an era? A mini-review. Hum Reprod 2005;20:2043-51.
5. Brown J, Farquhar C, Beck J, Boothroyd C, Hughes E. Clomiphene and anti-oestrogens for ovulation induction in PCOS. Cochrane Database Syst Rev 2009;4:CD002249.
6. Abu Hashim H. Management of women with clomifene citrate resistant polycystic ovary syndrome: An evidence based approach. In: Mukherjee S, editor. Polycystic Ovary Syndrome. ISBN 978-953-51-0094-2, InTech, 2012. p. 1-20. Available from: http://www.intechopen.com/articles/show/title/management-of-women-with clomifene-citrate-resistant-polycystic-ovary-syndrome-an-evidence-based-app. [Last accessed on 2012 Nov 13].
7. Overbeek A, Kuijper EA, Hendriks ML, Blankenstein MA, Ketel IJ, Twisk JW, *et al*. Clomiphene citrate resistance in relation to follicle-stimulating hormone receptor Ser680Ser-polymorphism in polycystic ovary syndrome. Hum Reprod 2009;24:2007-13.
8. Badawy A, El Nashar AB, El Totongy M. Clomiphene citrate plus *N*-acetyl cysteine versus clomiphene citrate for augmenting ovulation in the management of unexplained infertility: A randomized double-blind controlled trial. Fertil Steril 2006;86:647-50.
9. Rizk AY, Bedaiwy MA, Al-Inany HG. *N*-acetylcysteine is a novel adjuvant to clomiphene citrate in clomiphene citrate-resistant patients with polycystic ovary syndrome. Fertil Steril 2005;83:367-70.
10. Badawy A, State O, Abdelgawad S. *N*-Acetyl cysteine and clomiphene citrate for induction of ovulation in polycystic ovary syndrome: A cross-over trial. Acta Obstet Gynecol Scand 2007;86:218-22.
11. Borgstrom L, Kagedal B, Paulson O. Pharmacokinetics of *N*-acetylcysteine in man. Eur J Clin Pharmacol 1986;31:217-22.
12. Flughesu AM, Ciampelli M, Muzj G, Belosi C, Selvaggi L, Ayala GF. *N*-acetyl cysteine treatment improves insulin sensitivity in women with polycystic ovary syndrome. Fertil Steril 2002;77:1128-35.
13. Odetti P, Pesce C, Traverso N, Menini S, Mianeri EP, Cosso L. Comparative trial of *N*-acetyl cysteine, taurine and oxerutin on skin and kidney damage in long term experimental diabetes. Diabetes 2003;52:499-505.
14. Youssef G, Meguid Ali A, Alaa N, Makin B, Waly M, Abou-Setta A. *N*-acetyl-cysteine in anovulatory women: The impact of postcoital test. Middle East Fertil Soc J 2006;11:109-12.
15. Salehpour S, Tohidi M, Akhound MR, Amirzargar N. N Acetyl Cysteine, A novel remedy for poly cystic ovarian syndrome. Int J Fertil Steril 2009;3:66-73.
16. Elnashar A, Fahmy M, Mansour A, Ibrahim K. *N*-acetyl cysteine vs. metformin in treatment of clomiphene citrate–resistant polycystic ovary syndrome: A prospective randomized controlled study. Fertil Steril 2007;88:406-9.
17. Abu Hashim H, Anwar K, El-Fatah RA. *N*-acetyl cysteine plus clomiphene citrate versus metformin and clomiphene citrate in treatment of clomiphene-resistant polycystic ovary syndrome: A randomized controlled trial. J Womens Health (Larchmt) 2010;19:2043-8.

How to cite this article: Saha L, Kaur S, Saha PK. *N* -acetyl cysteine in clomiphene citrate resistant polycystic ovary syndrome: A review of reported outcomes. J Pharmacol Pharmacother 2013;4:187-91.
Source of Support: Nil, **Conflict of Interest:** None declared.