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Journal of Bioscience and Applied Research

www.jbaar.org

Breast cancer risk associated with genotype polymorphism of *COMT* gene in young women

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ABSTRAT

O- methelation mediated by COMT enzyme is an important mechanism for in activating Catechole Estrone (CE) which including 2- HE and 4- HE and transform it to 2-ME and 4-ME which act as anti- tumor genesis . Aim of this study was investigation the association between the polymorphism in *COMT* genotype and the breast cancer risk in (40) young patient's women aged between (20-39) years were diagnosed and confirmed with breast carcinoma at AL- Sadder hospital in Missan, and (40) healthy control women aged between (18-39) years in period between September -2009 to April -2010 Odds ratios(OR) and confidence interval (CI) were calculated in level significant $P < 0.05$. The statistical analysis showed no association between the breast cancer risk in young women and homozygous wild (Met/Met) genotype with an OR of 0.63(95%CI= 0.248 - 0.552), also with heterozygous (Val/Met) genotype with an OR of 0.93 (95%CI= 0.155 - 0.44), and when a combination (Met/Met + Met/Val) genotype with an OR of 0.78 (95%CI= 0.559-0.841), compared to homozygous mutant (Val/Val) genotype. No significant differences in frequency of low activity alleles between cases and controls, indicating the polymorphism as a single factor may not contribute to breast carcinogenesis in young women.

Abbreviations : COMT , Catechol -O- Methyl Transferase ; HE, Hydroxy Estrone ; ME, Methoxy Estrone; OR, Odds Ratios; CI, Confidence Interval ;Val, Valin ; Met, Methionen.

Keywords: *COMT* gene, Breast cancer, genotype, polymorphism, breast carcinoma

INTRODUCTION

The breast cancer is consider the second most commonest causes of cancer death and one of the most dreading disease of the women (**1**). No one known the exact causes of breast cancer but there are many risk factors that increase a person's chance of developing the disease .Estrogen exposure has been considered to be one of the main risk factor for breast cancer . One characteristic of estrogen is it's mitogenic action in hormone sensitive tissues such as uterus and breast (**2**).

Estrogen metabolite produce two catechol estrogens 2-hydroxyestradiol(2- HE) and 4- hydroxyestradiol (4- HE) have shown several biological effects , where the 4- hydroxyestradiol metabolite binds and activates the estrogen receptor with approximately the same affinity as estradiol However, the interaction with hormone receptor is markedly reduce for the 2-HE ,which therefore may possess a weaker hormonal potency as compared with the parent hormone estradiol . thus, promotes cell proliferation and carcinogenesis (**3**).

One of the major in activation pathways of 2- and 4-HE is through O-methelation of 2 and 4 – methoxy estradiol by enzyme Catechol - O - Methyle Transferase (COMT),where COMT activity is lightly present in liver and kidney ,and it is also found as significant levels in brain ,uterus in endomaterium and in the mammary gland (**4)**

Little is known about other factors implicated in breast carcinogenesis among young women . most of the current risk factors , including women menarche and menopause , age at the first full –time pregnancy as well as the number of parturition are indicators of cumulative estrogen exposure (**5**).

Recent studies hypothesized that the polymophisms in the gene coding for enzymes involved in estrogen metabolism may be predisposing for breast cancer.

The intention with present study was to investigate the association between the low –activity for *COMT* gene and risk of breast cancer in young women patients and compared with control population consisting of young healthy females.

MATERIAL AND METHODS

1- Collection of the samples:

Five ml of the peripheral blood samples were collected in sterilized tubes with EDTA from forty young female breast cancer patients aged between (20-39) years, and forty healthy female aged between (18-39)as control (blood donor). The breast cancer patients were diagnosed and confirmed with breast carcinoma at AL-Sadder hospital in Missan governorate between (September -2009 to April -2010) . Blood samples were transferred to the laboratory and kept directly in (-20C) until extraction of the genomic DNA.

2- Isolation and Estimation of DNA:

DNA was isolated by using modified Sambrook method (6) that was described by AL-Qurashi (7),where genomic DNA was obtained by congenital Phenol/Chloroform extraction followed by ethanol precipitation and storied at(-18C). The concentration of isolated DNA was calculated by spectrophotometric method using U.V- visible scanning spectrophotometer(Fig .1.).

3- Polymerase chain reaction:

The isolated DNA was assay by PCR with specific primers (*COMT* primers) forward 5- G CCCGCCTGCTGTCACC-3 and reveres 5-CTGAGGGGCCTGGTGATAGTG-3. The polymorphism of the COMT gene was studied according to protocol that described by (7)., after that the products were detected by electrophoresis in 2% agarose gel and *COMT* gene PCR fragment was 114bp.

PCR products was digested with Hsp 92II restriction enzyme and the digestion product was classified as homozygous wild type (50-64bp), Homozygous mutant type(50 or 64-114) and Heterozygous (50-114,69-114) alleles (Fig .2.).

THE RESULTS

The frequencies of the *COMT* genotype among the breast cancer patients and the control population are shown in Tab (1).

Statistical analysis by calculated Odds Ratios (OR)and P values ($P< 0.05$) revealed no association between the breast cancer risk and homozygous wild

(Met/Met) genotype with an OR of 0.63(95%CI= 0.248 - 0.552), also with heterozygous (Val/Met) genotype with an OR of 0.93 (95%CI= 0.155- 0.44) and when a combination (Met/Met + Met/Val) genotype with an OR of 0.78(95%CI= 0.559-0.841), compared to homozygous mutant (Val/Val) genotype.

Table (1) show the alleles frequencies of *COMT* genotype for young breast cancer patients and control population.

COMT genotypes	Control (%) no:40	Cases (%) no:40	OR	(95% CI =)
COMT Val /Val	**14 (35)**	**10 (25)**	**1.0**	
COMT *Val* / Met	**16 (40)**	**18 (45)**	**0.63**	**0. 248 - 0.552**
COMT Met /Met	**10(25)**	**12 (30)**	**0.93**	**0.155 –0.44**
COMT Met /Met + ***COMT*** Met /Val	**26 (65)**	**30(75)**	**0.78**	**0. 617- 0.883**

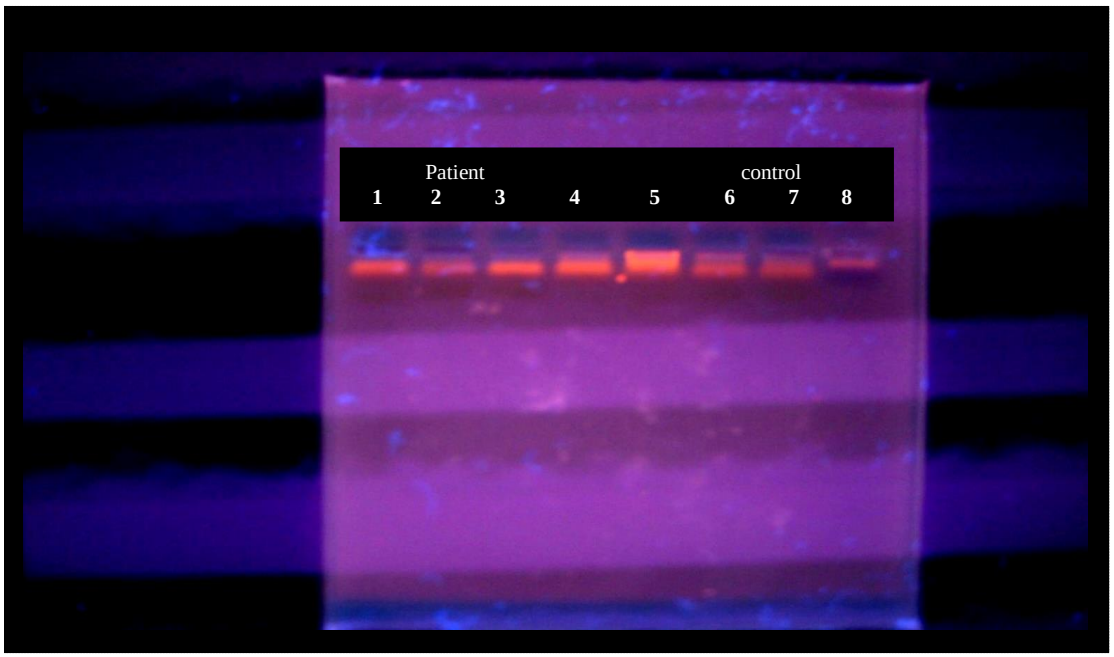

Fig .1. Agarose gel showing the molecular weight of DNA . Lanes (1,2,3,4,) the patients Lanes (5,6,7,8) normal women as control .

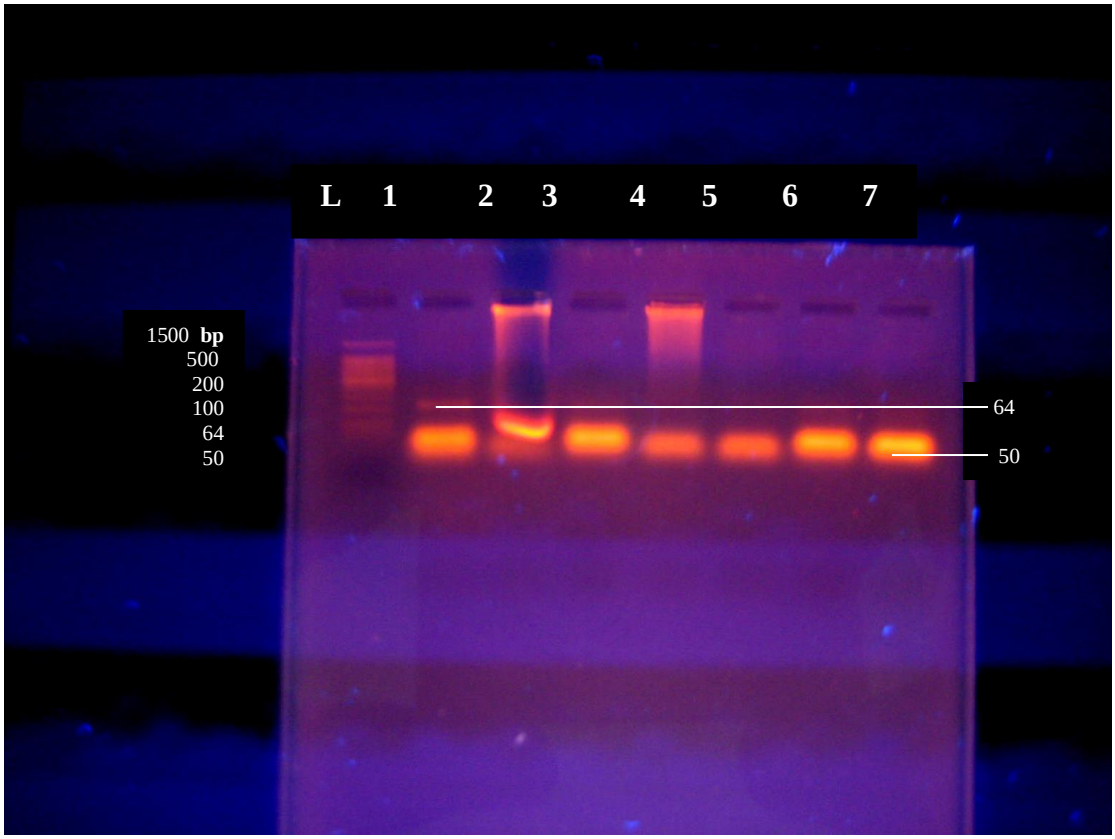

Fig .2. Hsp 92II restriction fragments of COMT alleles separated by agarose gel electrophoresis. L, Ladder .Lanes 3,6,7 show a homozygous low activity allele (Met/Met) , Lanes 4,5 show a homozygous high activity allele (Val/ Val) , and Lanes 1,2 the heterozygous pattern

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DISCUSSION

Analysis of genetic polymorphism in *COMT* gene exhibit no association between the low-activity alleles carriers (COMT Met) and breast cancer risk in young women .This results agreed with Millikan et al(8)they found no association between one or more copies of the low activity *COMT* allele and risk of breast cancer in study performed a population based case- control including 654 invasive breast cancer cases and 642 controls of both pre- and post menopausal women . Thompson *etal* (**9**) found no association between the low - activity (*COMT* Met) allele and breast cancer among pre menopausal individuals but , they found an inverse association in postmenopausal women .

Huang *etal* (**4**) found a significant association between low activity of COMT Met/Met genotype and breast cancer risk in postmenopausal women compared with pre menopausal women .While AL-Qurashi (**7**) found that the low activity of *COMT* Met allele have 3-4 fold resulting increase levels of circulating catechol estrogens and decreased formation of anti-tumorigenic , therefore ,the COMT Met allele may increase the breast cancer risk because decrease the ability to methylate and detoxify catechol estrogen.

Reduced of COMT activity may result enhanced cell proliferation and increased formation of free radicals as a consequence of a possible accumulation of 4-HE and furthermore, a decreased inhibition due to less synthesis of 2-ME2(**3**).

Anna et al (**10**) referred that decreased *COMT* activity might increase the risk of breast cancer through accumulation of(CE)which can causes oxidative to Quinines that react with DNA to form adduct this adduct can causes depuration leaving purine site ,which is the major type of genetic damage leading to mutation and genomic deletion during tumor genesis .

Body Mass Index (BMI) may be influence in breast cancer and this effect may be differ depending on menopausal status , where the postmenopausal adipose women have been reported to be at higher risk of breast cancer , while a decreased risk has been observed in pre menopausal women with high BMI (**11**). In this study we weren't collected the information about BMI because it isn't reported in the hospital .

In conclusion ,reduce (*COMT* Met) activity may in enhanced cell proliferation and increased accumulation of (4-HE) and decreased inhibition of angiogenesis due to loss synthesis of (2- ME). In this study we did not show increased in Angiogenesis of breast cancer risk associated with low –activity of COMT genotype.

REFERENCES

1. Laster ,SC. & Cotran ,R.S.(2003). The breast : Robbins pathologic of medicine Basis , *7th ED . Chicago Uni. P.Sch of medicine , P:23(1093).*

2. Zhu , B. T. and Conney , A.H. (1998) **.** Is 2-Methoxy estradiol an endogenous estrogen metabolite that inhibits mammary carcinogenesis ? *Cancer Res ; 58: 2269 - 2277.*

3. Bergman ,MJ.& Wingren,S. (2001). Catechol - O – Methyle Transferase *(COMT)*gene polymorphism and breast cancer risk in young women . *British Jol of Cancer 85 (6) :859 – 862* .

4. Huang , CH; Chern,H.D; Chang ,K.J; Hsu,S,M ; and Shen,Ch.Y.(1999). Breast cancer risk associated with genotype polymorphism of the estrogen – metabolizing Genes *CYP17, CYP1A1,* and *COMT* : Amultigenic study on cancer susceptibility.*Cancer Rech.Net.Taiwan University .59:4870 – 4875.*

5. Sprudle ,AB.; Hopper,JL.;Dite, GS.; and Chen,X.(2000). CYP17 promotor polymorphism and breast cancer in Australian women under age forty years . *J.Nat Cancer Inst 92: 1674 -1681*

6. Sam boork , J.; Fritsh, E.F and Maniatis , T. (1989). Molecular cloning , a laboratory manual. *2nd Ed . cold spring , harbor laboratory press. USA*.

7. AL- Qurashi ,K.J.(2008) . The role of *CYP1 B1* and *COMT* genes polymorphism as a risk factors of endometrial cancer in women .A thesis for Doctor of philosophy in biology , Bio-technology . *College of Science . Basrah University* .

8. Millikan,RC.; Pittman,GS. ; Duell,E. ; Newman,B. (1998). Catechol **-O -** Methyle Transferase and breast cancer risk . *Carcinogenesis 19:1943 -1947.*

9. Thompson , PA. ; Shields , PG.; Stone , A; Vena ,JE.; Marchall, JR. (1998). Genetic polymorphism in catechol -o- methyle tranferase , menopausal status and breast cancer risk . *Cancer Res58 :2107-2110.*

10. **Anna, H.**W. ; **Tseng , C. ; David, V.B.; and Mimi , C.V.(**2003) . Tea . Intake , COMT Genotype ,and Breast cancer in Asian –American women . *Amer . Asso . Cancer.Res (63): 7526-7529.*

11. Van den Brandt ,PA.; Spiegelman ,D. ; Yaun,SS.; Besson,L .; Folsom, AR.;Fraser,G.(2000). Pooled analysis of prospective cohort studies on height , weight , and breast cancer risk . *AM J Epidemiol 15:514 – 527.*