Metastasis continues to be the major cause of morbidity and mortality among cancer patients. Once the cancer has spread to distant sites the five-year survival rates for breast cancer drop from 100% to less than 25%. Therapeutic strategies mostly include excision of the primary tumor followed by radio-, and/or chemo which eradicates any remaining tumor cells that have escaped surgical removal. Although this approach is effective in many cases, reoccurrence of tumors and metastases is not uncommon. In fact, recurrent metastatic growth in breast cancer patients occurs years after the patient has been declared cancer free [93, 94].

Metastasis is the main complication in any type of cancer. The cascade of metastasis is quite complicated and systematic in its orientation regarding the involvement of cancer cells immune evasion, adhesion, invasion, motility, chemo attraction, cytoskeleton rearrangement, cell survival, gene rearrangement and unknown molecular factors. To prevent metastasis lot of research work is currently under way to understand the signalling cascade of the cells. Since metastasis is also genetically controlled event, several metastasis suppressor have been identified which are characterised by their reduced expression in highly metastatic compared with tumorigenic but poorly or non-metastatic, tumor cells. In addition, cohort studies have correlated the reduced expression of a metastasis suppressor to indicators of clinical aggressiveness, such as patient survival, development of nodal or distant metastases and tumor stage or grade [95, 96]. Although surgery to remove the primary tumor along with radiotherapy provides adequate control at the initial stage for most cancers, “metastasis” remains an inadequately addressed phase of cancer. Although metastasis requires many steps to complete the elimination of only one step in the chain would thwart the process, therefore restoration of metastasis suppressor gene expression in
metastatically competent cells might produce a clinical benefit in patients in which the metastasis process has not been completed. Analysis of the role of germ line polymorphism in metastatic will enlighten new aspects of this critically important cancer process. The fact that inheritance also seems to play a significant role in metastatic progression will provide an important clinical tool for prognosis and prevention of the most lethal aspect of cancer.

In our study we found no association of NME1 genotypes with breast cancer risk. However, an inverse association of breast cancer risk with both hetero (OR=0.32; 95\% CI= (0.17-0.58) as well as mutant genotypes (OR= 0.13; CI=0.04 - 0.40) of M KK 4 gene was found. When participants were classified on the basis of lymph node involvement a strong association between NME1 heterozygous genotype and presence of lymph node at time of diagnosis (OR=3.82; CI = (1.54 - 9.44) was found. On stratifying the cases on basis of ER, PR, and HER2 receptor status the associations could not be calculated and the relationship between cancer stage and genotypes was not found significant.

To the best of our knowledge our study is the first report suggesting an inverse association between M KK 4 polymorphism and breast cancer. The promoter influences the transcriptional activity of a gene and variations in promoter region influence its functions [84]. Change of 1304 T to G allele in the promoter region of the M KK 4 gene can increases its transcriptional activity which in turn increases the protein expression in cancerous tissue. Moreover, the level is more in patients harboring 1304G variant genotypes compared to patients with 1304TT genotype [85]. Therefore we might suggest that 1304T>G polymorphism contributes to decreased risk of breast cancer by increasing promoter activity. Our results corroborate the
findings of several other studies [84, 85]. Many studies have also supported that \textit{MKK4} has a role in tumor formation and development because it may act as a tumor suppressor gene [97-102]. The tumor suppressor role may also explain the decreased risk of breast cancer due to \textit{MKK4 polymorphism}.

In the study we did not find any association between \textit{NME1} variants and breast cancer risk. But we did find a strong association between \textit{EcoR1} polymorphism and lymph node involvement which is an indicator of aggressive disease behavior in patients with heterozygous genotype (OR=3.82; CI = 1.54 - 9.44). This implies that individuals with heterozygous condition are likely to have aggressive tumors. \textit{NME1} gene is down regulated in cells with high metastatic potential [90]. The available data also shows that lack of \textit{NME1} expression promotes metastasis [103] and its reduced expression is associated with aggressive characteristics like lymph node infiltration in several types of tumors including primary breast cancers [46]. The findings of our study therefore suggest inter individual difference in developing metastasis and support the fact that genetic polymorphisms of the metastasis suppressor genes can act as diagnostic as well as prognostic biomarkers which can have implications on cancer treatment as well as prevention of metastatic disease.

Our study suggests inverse association of \textit{MKK4} gene with breast cancer risk and elevated risk due to \textit{NME1} in subset of patients with lymph node involvement.

Small sample size is one of the main limitations of this study. Larger population-based, case control studies, as well as well-designed mechanistic studies, are warranted to validate our findings.