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Ph.D. Thesis Title: "Evaluating prognostic significance of Her-2/neu oncogene in the development of colorectal cancer"

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Abstract

Keywords: Colorectal Cancer(CRC), Her-2/neu polymorphism, Prognostic, SNP I655V(A/G), PCR-RFLP

Background: Human Epidermal Growth Factor Receptor 2 (HER-2/neu) has potent tumorigenic potential; Any mutation or overexpression of HER-2 can result into tumorigenesis (Coussens et al., 1985) as seen in breast/gastric/ovarian as well as in prostate cancer. It is generally accepted notion worldwide that colorectal cancer (CRC) results from various environmental and hereditary factors, inducing genetic changes in crucial genes involved in cell signaling and growth such as HER-2; overexpression and alterations in which are found in colorectal cancer patients. Controversies exist in the data of previous studies regarding prognostic significance of Her-2 overexpression and I655V SNP in the progression of colorectal cancer, reported from different parts of the world. Examining the impact of this overexpression and frequent HER-2 polymorphism in CRC in Indian patient population is still untouched and can be significant as racial or ethnic differences affect allelic frequency and polymorphism.

Methods: The present study was conducted to determine the prognostic significance of Her-2 overexpression and I655V SNP in in cases of CRC. The study was conducted after ethical approval of Institutional Ethics Committee and from collaborating institute. We conducted a patient based study analyzing 83 subjects who underwent colorectal cancer biopsy/colectomy. All samples were collected only after obtaining informed consent of the

study participants. A tissue biopsy/colectomy sample was collected from a single patient and divided into two halves; one half was stored in 1X PBS, pH 7.4 at -20º C which was used for isolation of genomic DNA and second half was stored in 10 % Buffered Formaldehyde at room temperature which was used for IHC. For molecular study, we also included 57 control subjects into the study and their blood was used for DNA isolation. Analysis of HER-2 polymorphism was done by PCR-RFLP technique.

Results & Conclusion: The mean age was found to be 55.9 years; median age was 56 years and mode age was 54 years with a range of 43 (30-73). Males constitute 63 (75.9%) and females constitute 20 (24.1%) of patient population. According to grade-wise distribution, 12 (14.45%) patients were of Grade I, 53 (63.85%) of Grade II and 18 (21.68%) were of Grade III. The overall positive expression for Her-2/neu was found in 34 (40.96%) cases. The pattern of Her-2/neu expression is significantly correlated with age; p value 0.000294 (*, $p < .05$) as maximum 41.97% (34/81) of cases were positive for Her-2/neu expression in the age group ≥45 years. 16.66%, 35.84% and 72.22% cases were positive for Her- 2/neu expression amongst grade I, grade II and grade III tumors respectively. The immuno-histochemical expression of Her-2/neu showed statistically significant correlation with histological grade p value 0.0045 (*, $p < .05$). We found out that out of 83 patients, 52 (62.65%) were of Homozygous Wild-type (A/A; Ile/Ile); 27 (32.53%) were of Heterozygous type (A/G; Ile/Val) and 4 (4.81%) were of Homozygous Mutant type (G/G; Val/Val). Allelic frequency of Ile (A) was found out to be 0.79 and that of Val (G) is 0.21 and were not significantly different from the control population. Fischer's Exact p-value obtained was 0.86. The average Val allele frequency among 59 populations globally is around 0.239 (Rajeevan, Soundararajan, Kidd, Pakstis, & Kidd, 2012) which indicate that the occurrence of I655V SNP is not involved in causation /progression of CRC in Indian patient population.