Hotspot mutations previously known to affect DNA contact or cause structural disruption were analyzed in a large cohort of colorectal cancer (CRC) patients. TP53 mutations are frequently observed in CRC, with an observed rate of 80%. Brown et al. found that TP53 mutations were strongly correlated with other gene mutations, such as MGMT (61% vs. 53%, p<0.0001) and TOPO2A (2.6% vs. 4.7%, p=0.002). These findings suggest that TP53 mutations may play a significant role in the pathogenesis of CRC.

**Background:** TP53 is a tumor suppressor gene that regulates cell cycle arrest and apoptosis. Mutations in TP53 are frequently observed in various cancers, including CRC. The presence of TP53 mutations has been associated with patient survival and response to therapy.

**Results:**

1. Characteristics of 3457 tumors included in the analysis

2. Mutations of TP53 and tumor location (a), patient gender (b), tumor histology (c), and primary vs. metastasis (d)

3. Structural distribution of TP53 mutations found in the CRC cohort. The X-axis represents the position on the TP53 gene, while the Y-axis shows the number of mutations found at a particular position. Intact mutations, known to affect DNA contact (R464, Q567Q), or cause structural disruption (R241Q, G245E, T249A, and R249S) are marked in red; additional nonsense mutations seen at high frequencies are marked in black.

4. Details on the most frequent TP53 mutations observed and the types of mutation

5. Molecular profile comparison of TP53 mutated and TP53 WT CRC tumors tested by IHC and ISH. A star indicates the differences are statistically significant (p<0.05) by Chi-square test.

6. Molecular profile comparison of TP53 mutated and TP53 WT CRC tumors tested by NextGen Sequencing. A star indicates the differences are statistically significant (p<0.05) by Chi-square test.

**Conclusions:**

- Analysis in a large cohort of colorectal cancer cohort reveals that TP53 mutations are seen in 62% of tumors and are associated with clinicopathological features such as tumor stage (left and right colon) and patient gender (higher in male and tumor histology (highest in adenocarcinoma)).

- Hotspot mutations previously known to affect DNA contact or cause structural disruption are seen at very high frequencies in our cohort; mutations in the DNA binding domain as well as nuclear export domain are also seen at high frequencies, which warrants further investigation. Future research will also include comparison with published TP53 datasets.

- While overall TP53 mutations are more likely to occur in males, mutations including R196 mutations are more likely to occur in right colon (p=0.008). While overall TP53 mutations are more likely to occur in similar frequency in primary tumors and metastases, R725 mutations are more likely to be found in metastatic tissue in the primary tumors (p<0.0001).

- Distinct molecular features associated with TP53 mutation in CRC included lower frequency of mutations in the DNA binding domain as well as nuclear export domain. Pathway activation was significantly lower in frequent mutations of PIK3CA, PTEN, and AKT1 and higher in microsatellite instability and amplification.

- Our findings suggest differential presence of therapeutic targets in CRC tumors based on TP53 mutation status.

**References:**