

NRAS GENE MUTATIONS IN COLORRECTAL CANCER

Introduction:

Definirlo como un grupo de células que crece incontrolablemente y que forma masas difusas que pueden distribuirse e invadir otros órganos del cuerpo. El cancer colorrectal son todos aquellos tumores que se asientan en el intestino grueso . Hoy en día constituye uno de los principales problemas de salud a nivel mundial. Además, no existe tratamiento optimo para el ya que los tratamientos existentes dañan también el resto de células del cuerpo. El único tratamiento curativo como tal es la cirugía pero no es valido para todos los tipos de canceres colorrectales y ademas no impide las recidivas.

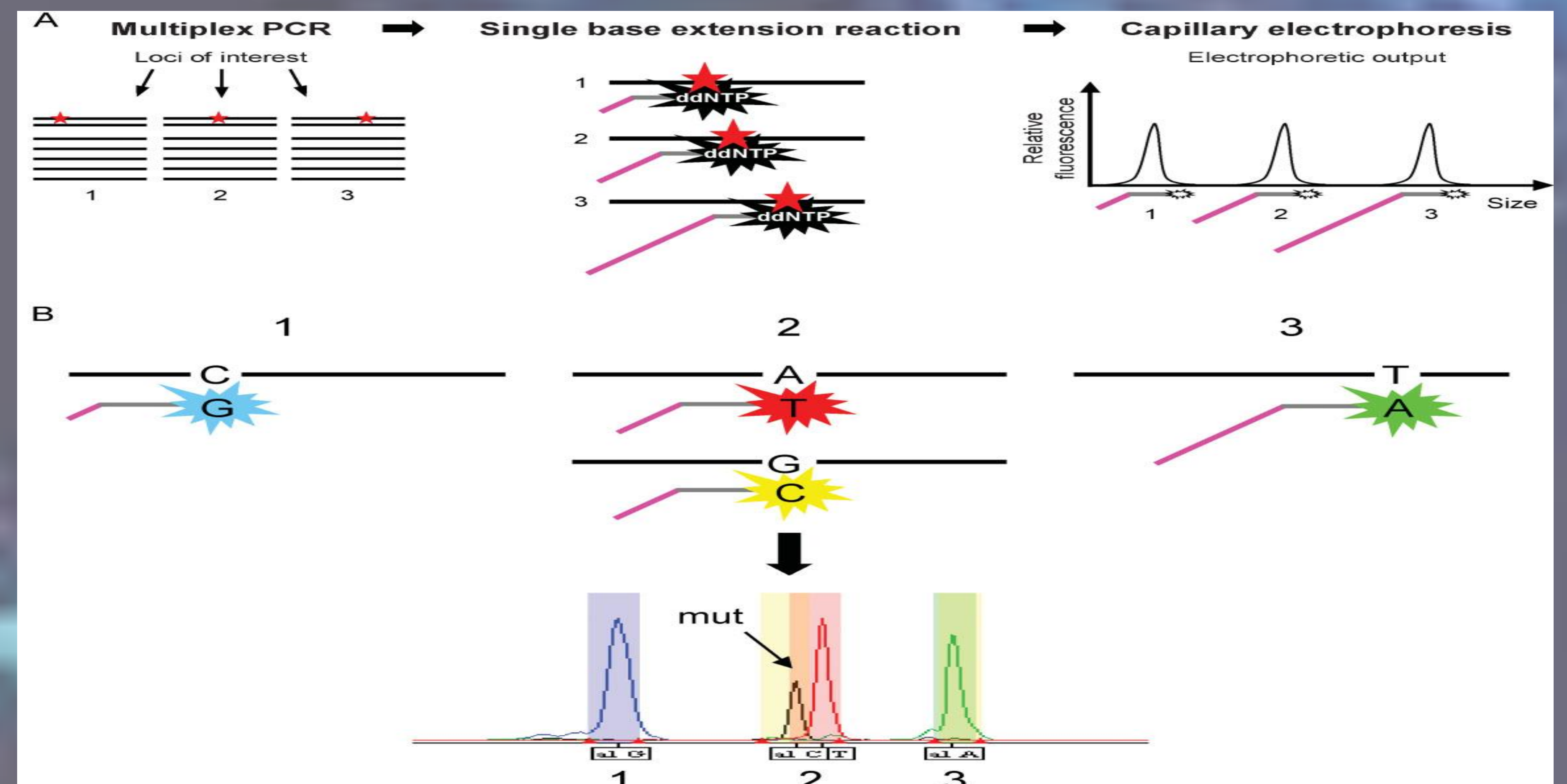
Por esta razon muchas de las investigaciones respecto al cancer se centran en la busqueda de su etiologia y de marcadores especificos que den lugar a tratamientos individualizados. Uno de estos marcadores es el gen RAS que afecta a nivel de una via importante para la division celular y su mutacion afecta en las opciones de tratamiento.

En este trabajo se analiza el gen N-Ras y su papel a nivel funcional en el cancer colorrectal y a nivel de respuesta al tratamiento.

Matherial and methods

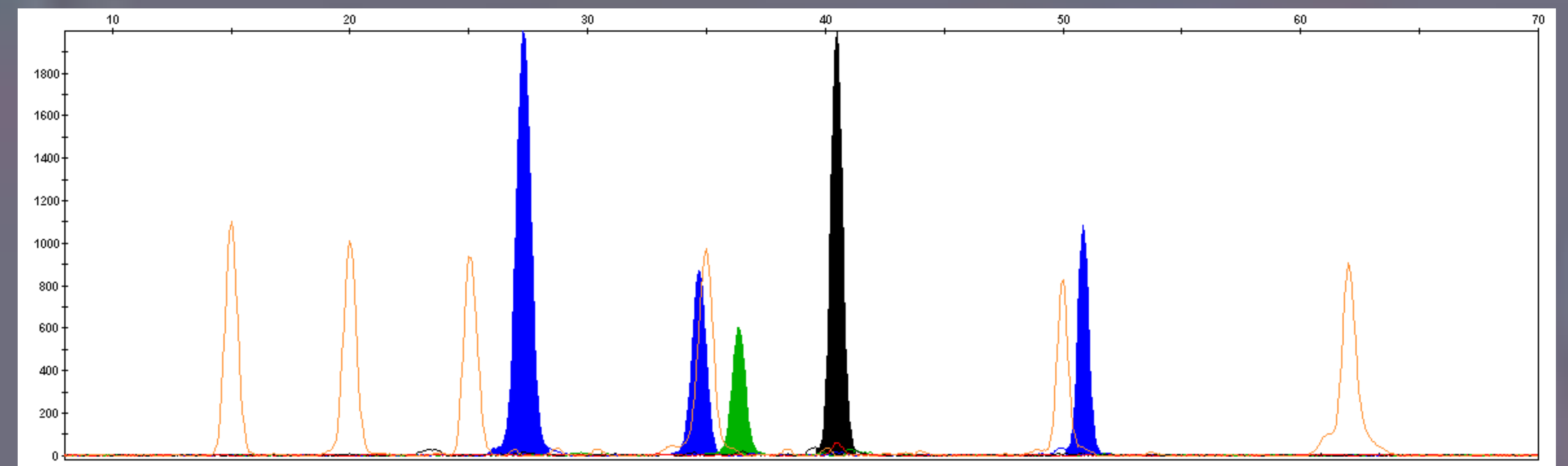
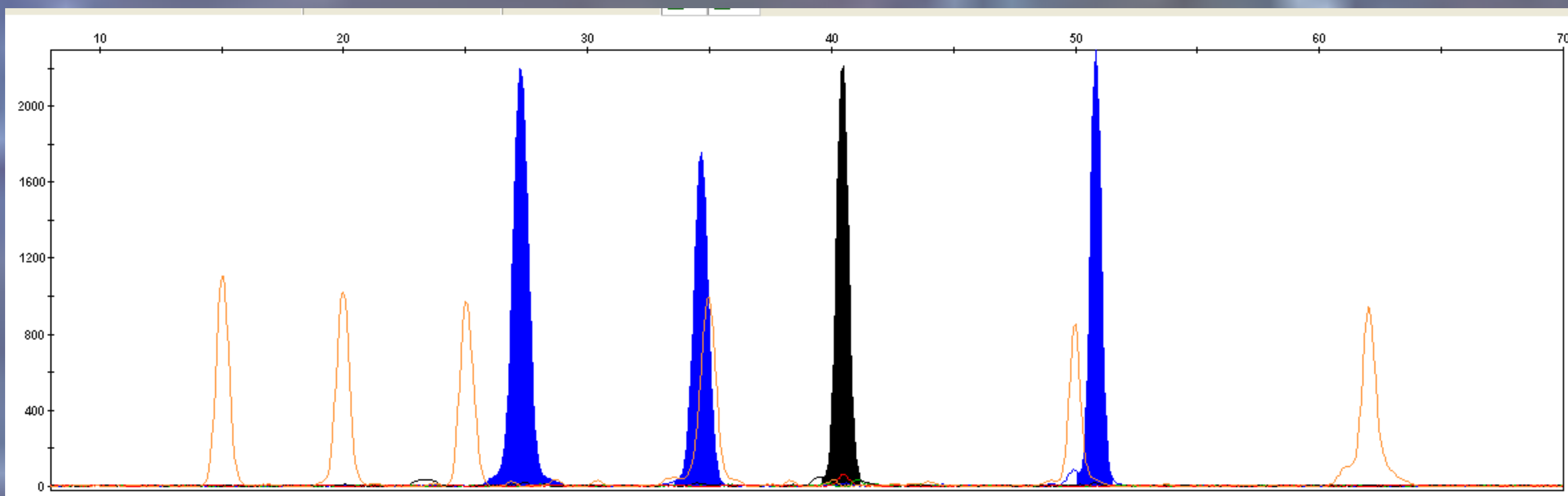
1. Sample selection: The sample was extracted from 84 different patients with colorectal cancer in different stages to analyze mutations produced in the N-ras gene in colorectal cancer
2. DNA extraction: the DNA was extracted from a sample of original tissue of each patient with the QIAmp DNA mini Kit.
3. Polimerase chain reaction (PCR): The PCR was performed for amplification of exons 2 and 3 being the most common sites of mutation in the NRAS. It was performed with KAPA Taq HotStart PCR kit
4. Gel electrophoresis and purification: the electrophoresis was performed to test the effectiveness of the reaction and the extent of the fragments obtained. Then samples were purified using a kit microcentrifuge column QIAquick Gel Extraction Kit.
5. Purification electrophoresis: this process was conducted to test the effectiveness of the purification process

6.Snapshot.: It is a single-base extension procces in which the primers and the nucleotides used allow us to distinguish each type of mutation. The nucleotides used are fluorescently labeled dideoxynucleotides each one with a different colorur. Besides the primers used were designed with different sizes to locate that mutation is shown



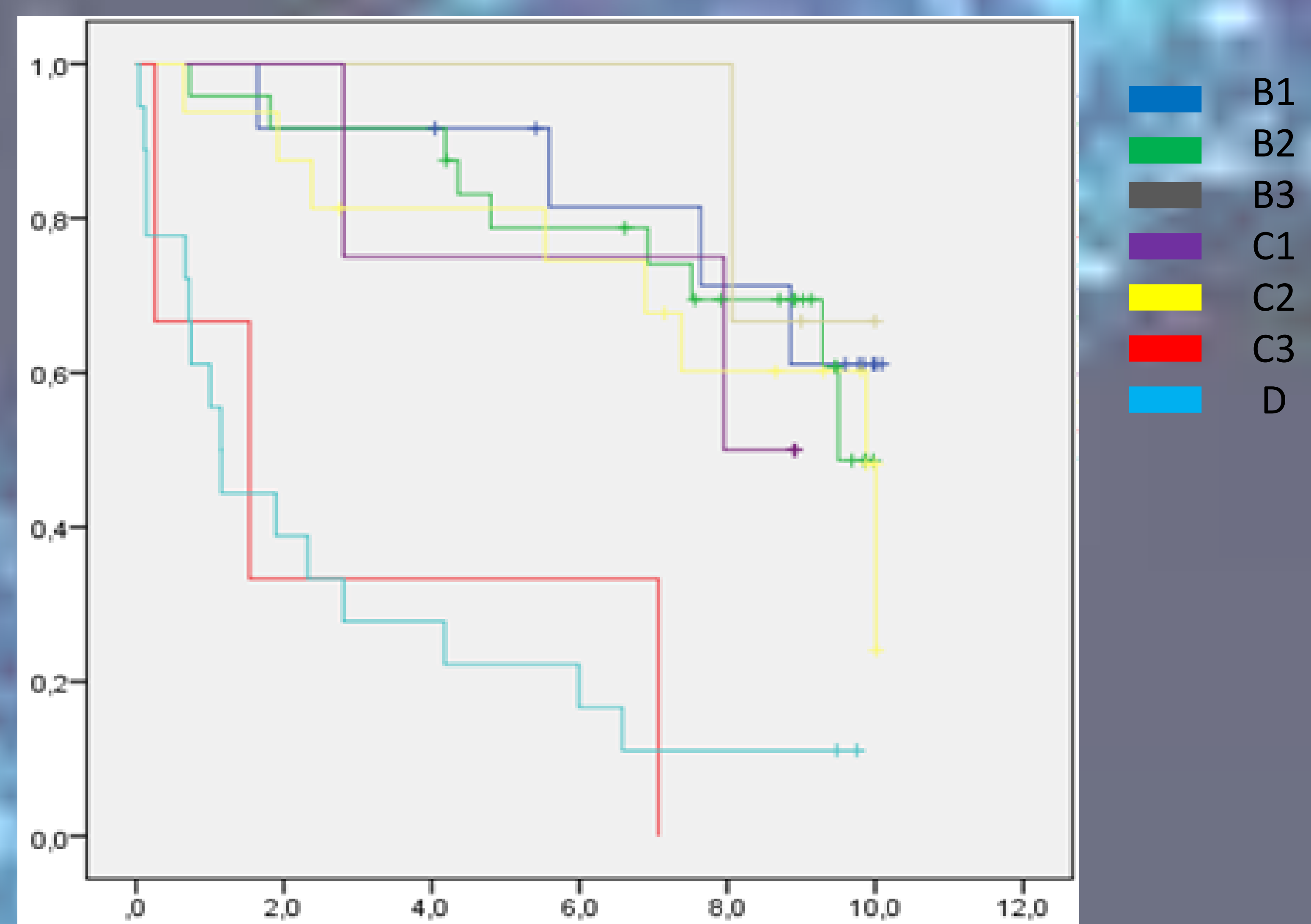
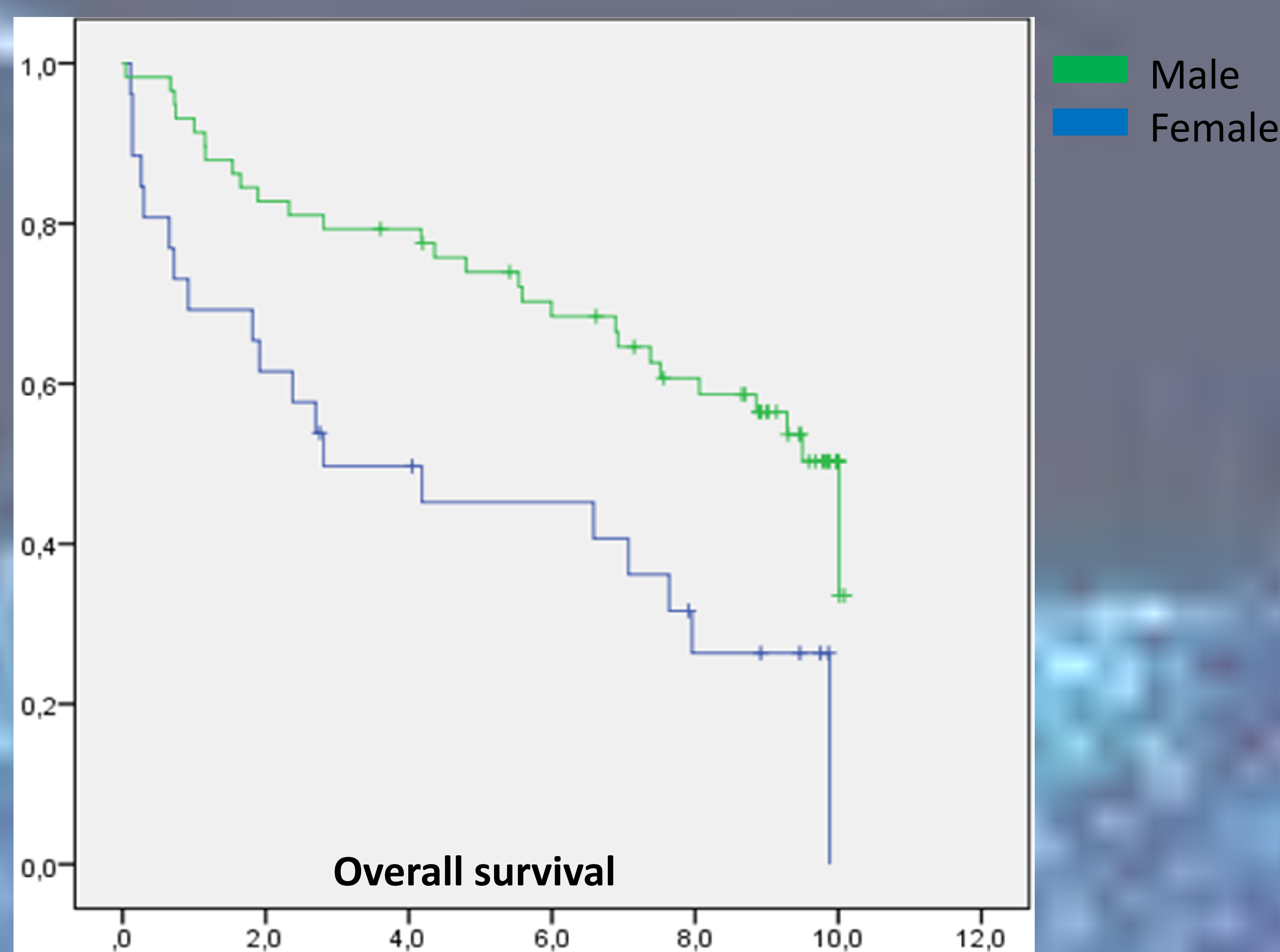
Results:

After mutation analysis 1.19% (N = 1) of patients showed mutations in the N-Ras gene while the remaining 98.81% did not have any mutation being considered wild type. This mutated patient had an age of 77 years and had a Dukes' stage D. In addition, presented various metastases and rank TNM was T3N2M1. The patient received chemotherapy in 2003 for other neoplasia. The 10-year survival of this patient is 0.7 years while the median survival of the remaining patients is 6 years.



The patient found had a mutation in the codon twelve of the exon two. This mutation was producing a change in the normal basis GGT to GAT. For this reason there is also a change the codon encoding the amino acid glycine (Gly) to aspartate (Asp).

Furthermore taking into account the clinical data of patients the survival of these were compared in function of the sex and the stage of the cancer



Discussion:

The findings of this study demonstrate that the mutation in the NRAS gene (1.19%) is uncommon. This information is consistent with information obtained in various studies that have studied these mutations and the genetic databases, but in contrast to other experiments our mutation appears in the 12 codon not in the 61. In colorectal cancer NRAS mutations seem to appear in more advanced stages than KRAS, appearing in early stages.

Moreover, ongoing research shows clear genotypic differences between tumor bearing animal models with altered NRAS and KRAS and proposes as a function of these genes and cell growth and apoptosis suppressing ability respectively.

Mutations in the gene NRAS are uncommon. Because of the low percentage of mutations in NRAS in colorectal cancer its final impact is still unknown