Cocoa and their products are sources of flavonoids and contribute in a significant proportion to the amount of dietary antioxidants. There is substantial experimental evidence that connects dietary flavonoids with the prevention of chronic diseases, such as cardiovascular diseases, diabetes or cancer. To further evaluate the role of cocoa flavonoids in cancer prevention, a functional genomic analysis was performed to study the effect of a polyphenolic cocoa extract (PCE) in the human breast cancer cell lines MCF-7 and SKBR3. Thus, changes in mRNA levels were determined using the specific Stress & Toxicity PathwayFinder™ RT2Profiler™ PCR Arrays. Upon incubation with PCE, 1 gene was upregulated and 7 were overexpressed in MCF-7 cells whereas 9 genes were overexpressed in SKBR3 cells (p < 0.05). Among the differentially expressed genes in both cell lines, CYP1A1 was chosen for further validation. The changes in expression for CYP1A1 upon incubation with PCE were confirmed at mRNA and protein levels. The roles of ER and HER2 in CYP1A1 induction were analyzed by means of the siRNA technology. We determined that ER triggered CYP1A1 induction through the AhR pathway in MCF-7 cells, whereas HER2 modulated CYP1A1 overexpression in response to cocoa extract in SKBR3 cells. To analyze a possible crosstalk between the different signalling pathways, AhR, ER; and HER2 proteins levels were determined by Western blot. The results showed that a crosstalk mechanism involving ER and AhR was activated upon incubation with PCE. Finally, the combination of PCE with tamoxifen was synergistic in both cell lines.

**Introduction:** Many epidemiological and preclinical studies suggest vitamin D compounds confer protection against colon cancer. We have reported that 1, 25-dihydroxyvitamin D3 (1,25 (OH) 2D3) inhibits the proliferation and promotes the differentiation of human colon cancer cells via the induction of the adhesion molecule E-cadherin and the antagonism of the Wnt/beta-catenin pathway. SPROUTY-2 (SPRY2) protein is an intracellular modulator of several growth factor tyrosine kinase receptors and therefore of cell growth and differentiation.

**Materials and Methods:** Human colon cancer SW480-ADH cells were cultured in DMEM. To knock-down gene expression, cells were infected with lentiviral particles containing a U6 promoter driving a shRNA targeting the respective RNA (Mission® shRNA lentiviral particles against human SPRY2, E-cadherin or scramble negative control, Sigma). To investigate SPRY2 expression in human colon tumours we considered sporadic cases if no clinical antecedents of Familial Adenomatous Polyposis were reported. Patients with clinical criteria of hereditary non-polyposis colorectal cancer (Amsterdam criteria) were excluded.

**Results:** SPRY2 expression in colon cancer cells was inhibited by 1,25 (OH) 2D3 through E-cadherin-dependent and -independent mechanisms. In turn, SPRY2 decreases both the basal and 1,25(OH)2D3-induced expression of E-cadherin, leading to the inhibition of the adhesive epithelial phenotype of colon cancer cells. Moreover, SPRY2 knock-down increases CDH1/E-cadherin expression and, reciprocally, CDH1/E-cadherin knock-down increases that of SPRY2. Consistently, SPRY2 and E-cadherin protein levels inversely correlate in colon cancer cell lines. In colon cancer patients, SPRY2 is up-regulated in undifferentiated high grade tumours and at the invasive front of low grade carcinomas and correlates inversely with that of E-cadherin.

**Conclusions:** Our data show that 1,25(OH)2D3 represses SPRY2, a negative regulator of E-cadherin that promotes dedifferentiation and invasion of colon cancer cells. These results reflect an unanticipated tumourigenic activity and potential of SPRY2 as a marker of malignancy, and a novel protective effect of 1,25 (OH) 2D3 against this neoplasia.