

Polyphenol compounds extracted from *Vaccinium myrtillus* bilberry has concentration-dependent antiproliferative effects on human prostate cancer cell lines

R. Bartoletti¹, C. Di Serio², F. Tarantini², A. Ungar², T. Di Vico¹, J. Durante¹, A. Tognarelli¹, L. Brizzi¹, M. Del Bubba³

1) Dept. of Translational Research and New Technologies University of Pisa, Italy

2) Dept. of Medical and Surgical Critical Area University of Florence, Italy

3) Dept. of Chemistry University of Florence, Italy

Aim of the study

Recently it has been demonstrated that spontaneous bilberry *Vaccinium myrtillus* is an excellent source of dietary polyphenols. Polyphenolic compounds exhibit several properties such as antioxidant, antimutagenic, antiinflammatory and anticancer activities. The anticancer activity of polyphenols is shown in a variety of human cancer cell lines both *in vitro* and *in vivo*, also in prostate cancer (PCa). PCa is recognized as one of the major medical problems in the male population and scientific evidences about polyphenols should be still demonstrated and well-conducted clinical studies are needed to clarify the efficacy of these molecules on PCa prevention and progression. Aim of this study is to investigate the effects of total polyphenols extracted from *Vaccinium myrtillus* on proliferation and apoptosis of three prostate cancer cell lines with different degree of malignancy compared with normal epithelial prostate cells.

Materials and the methods

Human prostate cancer LNCaP, DU145 and PC3 cell lines and normal human prostate epithelial cells (PrEC) were used to perform proliferation experiment and apoptosis analysis. Cell growth was assessed in condition of anchorage dependency and independency (Anchorage-independent soft agar) adding increasing amounts of bilberry extract in culture medium. Annexin V & Dead Cell Assay was performed utilizing Muse™ Cell Analyzer (Millipore) for apoptosis detection.

Results

With increasing concentration of *Vaccinium myrtillus* extract, increasing inhibition of cell proliferation in all tumor cell lines were observed, with different degrees of potency between cell lines, and having a low effect of inhibition on PrEC cells. In particular, higher concentrations of bilberry extract were necessary to obtain the same inhibitory effect on cell proliferation of androgen-independent DU145 and PC3 cells compared to androgen-dependent LNCaP cells. Growing LNCaP cell in soft agar, the treatment with *Vaccinium myrtillus* evidenced a reduction of growth capacity anchorage-dependent. Further, the bilberry extracts were evaluated for their ability to stimulate apoptosis. The flow cytometry revealed that *Vaccinium myrtillus* extract increased the total number of apoptotic cells in all carcinoma cell lines.



Conclusions

Our study provides data on the ability of these compounds to inhibit growth in PCa cells with a dose-dependent effect. Androgen-sensitive cell line LNCaP responds to treatment more than androgen-insensitive DU145 and PC3 while normal epithelial prostate cells are resistant. Antiproliferative activity appears to be mediated by pro-apoptotic mechanisms. These findings establish the potential value of *Vaccinium myrtillus* polyphenolic compounds as possible therapeutic agents against prostate cancer. Other investigations should furthermore focus on the bioavailable metabolites that are formed and their concentration in plasma and in prostate tissue.