CALIFORNIA STATE SCIENCE FAIR
2006 PROJECT SUMMARY

Name(s)

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Project Number

S1406

Project Title

Anti-Tumor Action of Indole-3-Carbinol by Suppression of NF-kappa B Activation

Abstract

Objectives/Goals

Indole-3-carbinol (I3C) is a compound occurring naturally in Brassica species vegetables (e.g., cabbage, broccoli). In vitro, I3C has been shown to suppress the proliferation of various tumor cells, and, in vivo, it has suppressed tumorigenesis. This project examines a possible mechanism of anti-cancer action at the molecular level for I3C by interference with the nuclear factor-kappaB (NF-kB) activation pathway that is a critical determinant of the inflammatory response, immune response, and apoptosis.

Methods/Materials

Two cell lines (Jurkat and GTL-16) were pretreated with I3C (10, 30, 60, or 100 µM) in culture media (DMEM or RPMI 1640) for 24 hours at 37°C (5% CO2) in T75 culture flasks. Three different activators of the NF-kB pathway were added individually to separate cell cultures: 20 ng/mL TNF-alpha for 5 minutes; 100 ng/mL PMA for 10 minutes; or 10 µg/mL LPS for 1 hour. Cell extracts were prepared per extraction kit manufacturers instructions. Sample measurements were performed with three ELISA assays: NF-kB; IkBa-Phospho-specific; and IkBa-Total per manufactures instructions. ELISA plate reading and data analysis was preformed on Molecular Devices reader and software.

Results

Through ELISA measurement of cell extracts for: 1) NF-kB, 2) IkBa-[pS32], and 3) IkBa-Total, this study found that I3C suppressed NF-kB activation by various agents (TNF-a, PMA, LPS) in two cell lines (Jurkat and GTL-16). NF-kB inhibition correlated with suppression of IkBa phosphorylation, IkBa degradation, and NF-kB nuclear translocation in a dose dependent manner. For Jurkat cells treated with 20 ng/mL TNF-a for 5 minutes, the 100 µM I3C dose produced: 1) NF-kB values of 150 ng/mL (vs 327 ng/mL with no I3C); Phospho-IkBa values of 207 units/mL (vs 336 units/mL with no I3C); and Total IkBa values of 3.84 ng/mL (vs 2.19 ng/mL with no I3C).

Conclusions/Discussion

Overall, the results demonstrate that I3C is an effective inhibitor of NF-kB activation, which may explain its antiproliferative, proapoptotic, and antimetastatic effects. Studies that monitor individuals diets and their health have found links between certain types of food and cancer risk. To explain the connection how certain foods protect against cancer, it is necessary to understand the process at a molecular level. Continued research is uncovering how crucial cancer genes can be influenced by compounds in the things we eat.

Summary Statement

Indole-3-Carbinol found in Brassica sp. vegetables mediates anti-tumor activity through NF-kappa B modulation in a dose dependent manner.

Help Received

Used laboratory equipment at BioSource; Dr. John DeSimone provided training and guidance in ELISA testing and cell culture handling.