P017 Withaferin a is a potent inducer of the Nrf2-mediated environmental stress response

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Withania somnifera (WS) is a botanical that has been long used in traditional Indian medicine. Withaferin A (WA) isolated from WS shows therapeutic and preventive activity in experimental models, but the mechanism of action is less understood. We hypothesized that WA induces Nrf2 signaling, resulting in protection of cells and organisms against chemical stresses. Human mammary epithelial MCF10A cells or wild-type (WT), Nrf2-disrupted, Keap1-disrupted, Keap1/Nrf2 double-disrupted mouse embryonic fibroblasts (MEF) were treated with graded WA doses. Enzyme expression was quantified by qRT-PCR and western blot**.** Nrf2 target gene induction was observed in MCF10A (CD= 80 nM), WT-MEF (CD= 200 nM) and Keap1-disrupted MEF compared to Nrf2-disrupted and Nrf2/Keap1 double-disrupted MEF. WA is more potent than sulforaphane (CD=1.5 μM) in inducing NQO1 transcripts in MCF10A. WT and Nrf2-disrupted C57BL/6J mice were gavaged with DMSO or 7 mg/kg WA. NQO1 transcript induction was observed in liver, small intestine, lung, colon and brain of WA-treated WT-mice, but not in Nrf2-disrupted mice. Repeated WA dosing produced increased cytoprotective gene expression in liver. The protective role of WA was assessed by challenging mice with 300 mg/kg acetaminophen (i.p). WA-administered WT-mice were protected from acetaminophen hepatotoxicity as evidenced by attenuated serum ALT and liver damage. Thus, WA is a potent Nrf2 inducer that protects cells and organisms against toxicity in an Nrf2-dependent, Keap1-independent mechanism and may be a useful chemopreventive agent. Support: NIH R01 CA94076 and Breast Cancer Research Foundation.