

## ABSTRACT

### Chemopreventive and anticancer potential of Jamaican natural and synthetic compounds

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Given the high endemism in biodiversity within Jamaica, this thesis undertook to investigate isolated natural products (and some synthetic ones) for their biological worth using two screens, towards estimating chemopreventive and chemotherapeutic potential. This thesis reports for the first time inhibitory properties of Jamaican natural and synthetic compounds on CYP1 family of enzymes (CYPs1A1, 1A2 and 1B1) and their impact on the viability of cancer (HT29, HepG, and MCF-7) and normal (CCD18 Co) cell lines.

Bioactivity screens were carried out on 22 compounds isolated from terrestrial (*Amyris plumieri*, *Castella macrophylla*, *Clusia flava* and *Garcinia humilis*), marine (*Cymopolia barbata*) and synthetic sources. The 22 compounds belonged to 8 families; chromene amides (CAs), coumarins (C), polyisoprenylated benzophenones (PBPs), prenylated bromohydroquinones (PBQs), quassinoids (Qs), xanthenes (Xs) along with synthetic amides (SAs) and quinoline (SQ). IC<sub>50</sub>s were determined against the activities of CYP1 family using a fluorometric assay that was initially optimized using known inhibitor standards and derived Michealis constants. For those compounds that displayed moderate-potent inhibition (IC<sub>50</sub> < 10 μM), further kinetic studies were carried out to determine the nature of inhibition along with their inhibition constants K<sub>i</sub>s. Also, investigations against other drug metabolising enzymes; CYPs2C19, 2D6 and 3A4 were carried out. All compounds were then examined for cytotoxicity potential using the MTS assay.

Of the 22 compounds investigated, 6 promising lead compounds from 6 classes were identified; 6-Methoxy-3-methyl-N—(pyridin-2-yl)benzofuran-2-carboxamide (SA1), 7-hydrocycymopolone (PBQ2), chromene amide with a n-butanamide side group (CA3), brasixanthone (X2), (-)- Glaucarubolone glucoside (Q3) and scopoletin (C1). Three of these compounds, SA1, PBQ2 and CA3 exhibited dual bioactivity properties (CYP1 inhibition and cytotoxicity towards one or more of the cell lines). SA1 was potent and selective towards CYP1B1 (IC<sub>50</sub> of 0.28 ± 0.2 μM) in an uncompetitive manner (K<sub>i</sub> of 0.068 ± 0.009 μM) and impacted the viability of cancer colon cells (IC<sub>50</sub> of 31.83 ± 3.4 μM) comparable to the chemotherapeutic drug fluorouracil (IC<sub>50</sub> of 23.50 ± 1.12 μM). However, while fluorouracil impacted normal colon cells (IC<sub>50</sub> of 55.51 ± 3.71 μM), SA1 favourably did not impact these cells (IC<sub>50</sub> > 60μM). PBQ2 inhibited not one but both CYPs1A1 (IC<sub>50</sub> 0.93 ± 0.3 μM) and 1B1 (IC<sub>50</sub> 0.14 ± 0.0 μM) at high potency in a noncompetitive manner (K<sub>i</sub> of 0.84± 0.07 μM for CYP1A1 and 4.7\*10<sup>-3</sup> ± 5.1\*10<sup>-4</sup> μM for CYP1B1).

Further, PBQ2 selectively impacted colon cells and the obtained IC<sub>50</sub> was parallel to that of fluorouracil for cancer (IC<sub>50</sub> of 19.82 ± 0.46 μM) and normal (IC<sub>50</sub> of 55.65 ± 3.28 μM) colon cells.

CA3 exhibited high selectivity towards CYP1A1 with an IC<sub>50</sub> value of 2.43 ± 0.62 μM and impacted breast cancer cells although potency was not comparable to tamoxifen. Structure activity relationship studies inferred the vital role played by the aliphatic branched side chains (under 7 carbons) in binding CYPs1A2 and 1B1 and as such provided key information when searching for other CAs with potential for chemoprevention and chemoprotection.

The other three compounds that demonstrated promising bioactivity were (-)-glaucarubolone glucoside (Q3), scopoletin (C1), and brasixanthone (X2). Q3 was more effective in reducing breast cancer cell viability than known chemotherapeutic drug tamoxifen (IC<sub>50</sub> of 8.65 ± 1.11 μM for Q3 vs. 17.28 ± 0.06 μM for tamoxifen during 24 hr incubations) while more recent studies in the laboratory has proven cytotoxicity at the nano molar range for 48 and 72 hour incubations. Further, Q3 was shown to induce early apoptosis in breast cancer cells using Annexin V-FITC dye assay. Like SA1, C1 selectively reduced the viability of cancer colon cells (IC<sub>50</sub> of 19.28 ± 0.35 μM) comparably to known chemotherapeutic drug, fluorouracil while showing no impact towards normal colon cells unlike fluorouracil. X2 selectively and potently inhibited CYP1A1 (0.13 ± 0.00 μM) in an uncompetive manner (K<sub>i</sub> of 0.08 ± 0.01 μM).

Further research on compounds described whose bioactivity has been identified in this thesis will likely provide useful therapeutics with implications in the regime of chemoprevention and chemotherapy.

**Keywords:** Simone Badal McCreath; CYP450, Chemoprevention, Anticancer, Jamaica natural products