SYNTHESIS AND ANTICANCER ACTIVITY OF RESVERATROL DERIVATIVES

KOTAGIRI SARITA[1], ARCHANA SWAMY[1], SREEDHAR CHANDANA[2], KSHAMA DEVI[3]

[1]Gautham College of Pharmacy, RT Nagar, Bangalore, Karnataka.

[2]Acharya & B.M. Reddy College of Pharmacy, Bangalore, Karnatka.

[3]Al-Ameen College of Pharmacy, Bangalore, Karnataka

ABSTRACT

Few derivatives of Resveratrol were synthesized using appropriate synthetic route. The structures of these compounds were established by means of TLC, IR and ^{1}H-NMR and elemental analysis. All the compounds were evaluated for anticancer activity. Compounds showed significant activity.

1.INTRODUCTION

Resveratrol is a phytoalexin found in grapes and other food products like peanuts,mulberry (Siemann and Creasy,1992). It is known to have cancer chemopreventive activity (Jang,1997) and is found to decrease the tumour growth in rat tumour model (Costelli,1999). According to ciolino and Yen (1999) Resveratrol can inhibit aryl hydrocarbon induced carcinogenesis in animals. Mannila (1993) reported that its antileukemic activity on mice leukemia system L-1210 is very less as compared to other stilbenes present in *Picea abies bark.*

The rationality behind the design of semisynthetic derivatives/analogues of Resveratrol is based on the fact that, stilbene compounds are moderately stable over a period of time (Trela and Waterhouse,1996; Pezet and Cvenat,1996). It is speculated that the incorporation of acetyl, methyl and benzoyl group on the phenolic –OH group of Resveratrol, could prevent its oxidation to air and light reaction. Also, these group incorporation eventually produce ester/ether and these may function not only as pro-drug but also promote better effective toxicity.

Thus derivatization of Resveratrol was found undertaken with different reagents. The Resveratrol required for these reactions was obtained partly in pure form and the rest was purified from 20% W/W sample.

2.EXPERIMENTAL

The anticancer activity of the synthesized compounds was determined by Brine shrimp lethality Bioassay method (Meyer,1982).

Preparation of Resveratrol triacetate:

A drop of concentrated sulpuric acid is added to Resveratrol 1g in acetic anhydride 15 ml and the reaction mixture is stirred for three hours. The completion of the reaction is confirmed by TLC. The mixure is washed with distilled water 100ml and extracted with ethyl acetate 200 ml. The Ethyl acetate extract is dried over anhydrous sodium sulphate and the solvent is evaporated to give light yellowish brown powder.

Preparation of Trimethylether of Resveratrol:

Potassium carbonate 3.8 g is added to the solution of Resveratrol 1 g in dimethyl formamide 10 ml and stirred for half an hour at 0°C. To this reaction mixture, added the solution of methyl iodide 2.6 g in dimethyl formamide 10 ml dropwise over the period of half an hour. Stirred the reaction contents for five hours. The reaction is monitored by TLC. The reaction mixture is washed with water 300 ml and extracted with ethyl acetate (3 × 100 ml). The ethyl acetate extracts are pooled together and dried over anhydrous sodium sulphate. The solvent is removed to recover a brown solid.

Preparation of Resveratrol tribenzoate:

Benzoyl chloride 1g is added to the solution of resveratrol 1g in pyridine 10 ml dropwise over the period of half an hour. The reaction mixture is stirred for six hours. The completion of reaction is confirmed by TLC. The reaction mass is washed with 5% sodium carbonate 200 ml, extracted with ethyl acetate. The ethyl acetate extract is dried over anhydrous sodium sulphate and the solvent is evaporated to give a dark brown solid.

3.RESULTS AND DISCUSSION

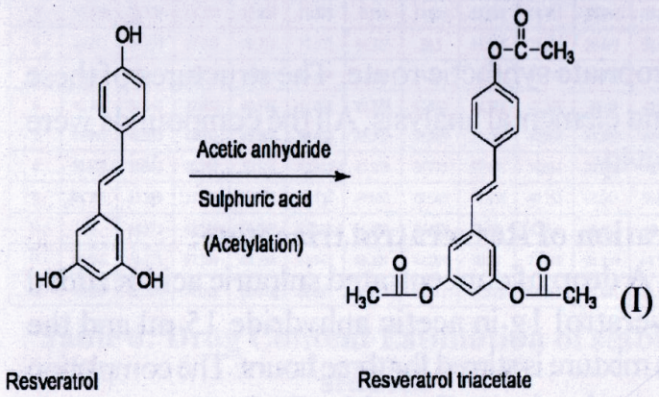
From the table no. 3 it is clear that pure Resveratrol is more potent than the derivatives in existing toxicity to shrimps. However, out of the three derivatives

***Corresponding author**

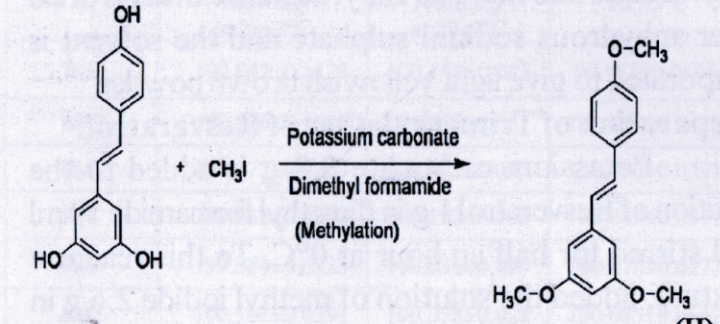
Email: gauthamcollege@gmail.com

of Resveratrol, only resveratrol tribenzoate has shown better cell killing activity. However, they were in no way superior to Resveratrol in eliciting such activity, suggesting that the free phenolic group in Reveratrol may be useful chemical moiety in exhibiting the cytotoxic effect.

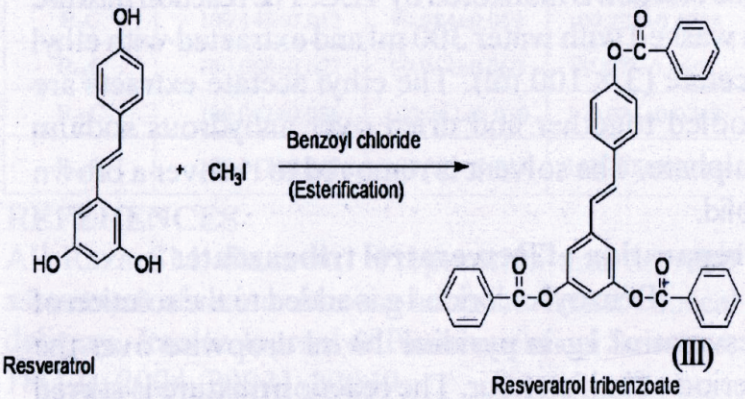
SCHEME OF SYNTHESIS:



Resveratrol — Resveratrol triacetate



Resveratrol — Trimethylether Resveratrol



Resveratrol — Resveratrol tribenzoate

Table No. 1: Physicochemical properties of Derivatives:

Compound	Molecular formula	Molecular Weight (g)	M.P. (°C)*	% yield (%)	R_f**
I	$C_{20}H_{18}O_6$	354	111-112	77	0.76
II	$C_{17}H_{18}O_3$	270	56	84.74	0.88
III	$C_{35}H_{24}O_6$	540	161	53.47	0.98

*Uncorrected, **Solvent system: Benzene : Ethyl acetate (8:2)

Table No. 2: Spectral Data of synthesized compounds:

Compd.	IR (cm^{-1})	^{1}NMR (, ppm)
I	1767 (C=O ester str.), 1199 (C-O str.), 912 (C-H bend.), 2926, 2853 (Ar C-H str.), 1371 (C-H str.), 1621 (Ar. C-C str.)	2.2 (s, 9H, 3 × CH_3), 7.1 (d, 2H, 2 × CH), 6.74-7.40 (m, 7H, Ar-CH)
II	1253 (C-O str.), 790, 762 (C-H bend.), 2837 (Methyl C-H str.), 681 (C-C bend), 2951 (Ar C-H str.), 1623 (Ar. C-C str.)	3.74 (s, 9H, 3 × CH_3), 7.2 (d, 2H, 2 × CH), 6.52-7.40 (m, 7H, Ar-CH)
III	1736 (C=O ester str.), 708 (C-H bend.), 1338 (C-H str.), 1199 (C-O str.), 2964 (Ar C-H str.), 1626 (Ar. C-C str.)	7.11 (d, 2H, 2 × CH), 6.89-7.40 (m, 22H, Ar-CH)

Table No. 3 Results of Brine Shrimp Lithality bioassay

Sr. No.	Drug	% death after 24 h						LC 50 (µg)	95% confidence interval
		1 µg	5 µg	10 µg	20 µg	100 µg	1000 µg		
1	Cyclophosphamide	10.0	55.0	60.0	70.0	100.0	100.0	3	4.2-1.4
2	Pure Resveratrol	10.0	50.0	70.0	80.0	100.0	100.0	5	7.6-2.4
3	Resveratrol triacetate	10.0	25.0	35.01	50.0	100.0	100.0	20	31.2-10.2
4	Resveratrol tribenzoate	10.0	20.0	50.0	80.0	100.0	100.0	10	15.6-5.0
5	Resveratrol trimethylether	0.0	20.0	30.0	70.0	100.0	100.0	16	23.8-7.8

Controls (Tween 80 suspension) showed 0 % death after 24 h.

REFERENCES

Ciolino HP and Yen GC, Inhibition of Aryl hydrocarbon-induced cytochrome P-450 IAI enzyme activity and CYPIAI expression by Resveratrol, Mol.Pharmacol., 56, 1999, 760-767.

Costelli P, Carbo N, Francesco MB, Francisco JLS and Joseph MA, Resveratrol, a natural product present in wine decreases tumour growth in a rat tumour model, Biochem.Biophys. Res.Commun., 254, 1999, 739-743.

Jang M, Lining C, Udeani OH, Karla VS, Christopher, Fong WWB, Cancer Chemoprotective activity of Resveratrol, a Natural Product derived from grapes, Sciences, 275, 1997, 218-220.

Mannila E, Talvitie A and Erikki K, Antileukemic compounds derived from stilbenes in Picea abies bark, Phytochemistry, 33, 1993, 813-816.

Meyer BN, Ferrigni NR, Putnam JE, Jacobsen LB, Nicholas DE and McLaughlin, Brine Shrimp: A convenient general biassay for active plant constituents, Planta Medica, 45, 1982, 31-34.

Pezet R and Cvenat P, Resveratrol in wine, Extraction from Gamay grapes, Am.J.Enol.Vitic., 47, 1996, 287-290.

Siemann EH and Creasy LL, Concentration of the phytoalexin Resveratrol in wine, Am.J. Enol.Vitic., 43, 1992, 49-52.

Trela B C and Waterhouse A L, Resveratrol: Isomeric molar absorptivities and stability, J. Agric.Food Chem., 44, 1996, 1253-1257.