RESEARCH REPORT

Bioactive Extracts from *Senecio lyratus*

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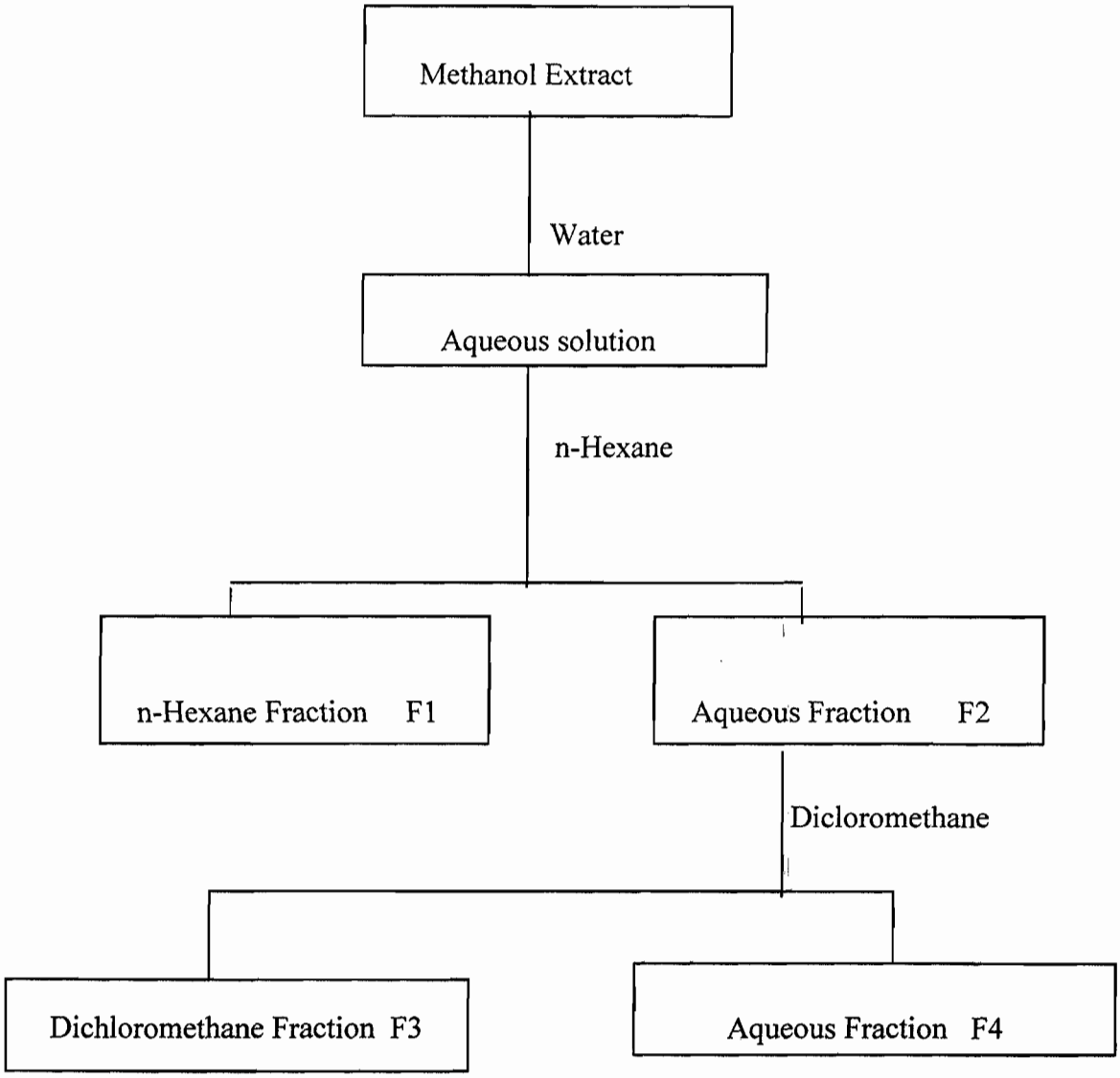
Dichloromethane, methanol and aqueous fractions from dry methanol extracts of *Senecio lyratus* showed activity when assayed on brine shrimp (*Artemia salina*). Except the methanol extract, the other two also showed activity when tested on *Escherichia coli* (Gram-negative bacteria) and *Staphyllococcus aureus* (Gram-positive bacteria). Gravity column chromatography of the aqueous fraction led to the isolation of a pure yellowish amorphous compound, Rf 0.62 (uncharacterized) when developed with n-hexane, ethyl acetate (40:60). The compound exhibited acute lethal effects on *Aspergillus niger, Bacillus subtilis, Micrococcus luteus, S. aureus and E. coli.*

Senecio lyratus belongs to the family *Compositae* (Agnew and Shirley, 1994) and is an ethnomedically important plant. It is distributed in altitudes between 1500m and 2760m above sea level and in Meru district, Kenya it is used for treatment of wounds and as an emetic (Kokwaro, 1976). The chief chemical components of the genus are pyrrolizidine alkaloids (Robins and Fortschr, 1982).

The purpose of this study therefore was to extract dry ground aeriol parts of *S. lyratus* using cold methano and to partition aqueous solution of the dried extract using n-hexane and dichloromethane and to evaluate the biological activity of all the fractions using brine shrimp, *Escherichia coli, Staphyllococcus aureus, Aspergillus niger, Bacillus subtilis* and *Micrococcus luteus.*

Aerial parts of *S. lyratus* were collected from Kericho district, Kenya. A voucher specimen is deposited in the herbarium of the University of Nairobi. The flow chart below summarizes the extraction and partitioning processes.

Isolation of pure compound from aqueous fraction (F4) concentrate



The concentrate (1.3g) was pre-absorbed on silica gel and subjected to a column packed under n-hexane with silica gel. Elution of the column with n-hexane and ethyl acetate (40:60) led to the isolation of a pure, UV sensitive compound with R_f 0.62 (eluent: n-hexane and ethyl acetate 70.30).

Brine shrimp lethality test was performed according McLauglin et al. 1989. *A. salina* (brine shrimp) were hatched into nauplii (nymphs) after 48 hours in artificial "sea water" in the dark and allowed to swim to a lighted area through perforations in the hatching vessel. Dichloromethane (F3, n-hexane (F1) and aqueous fraction (F4) were subjected to this test. The test solutions were prepared at concentrations of 10, 25, 50 and 100 ppm (1ml each) in triplicate in 2ml dram vials in acetone. The acetone was allowed to evaporate and replaced with the "sea water" containing nauplii (10 each). The number of nauplii still alive were

monitored after 24 hours. The control experiments were performed in vials with no samples. The data was processed in a simple program on a personal computer to estimate LC_{50} values with 95% confidence intervals for statistically significant comparison of potencies. Results are summarized in Table 1.

E. coli and *S. aureus* was each spread on a sterile petri dish smeared with agar (growth medium). Sterile blanks were stained with 100 ppm of the pure compound, dichloromethane fraction, n-hexane fraction and aqueous fraction (F4) and then transferred to the center of the agar plate before being incubated for 24 hours at 37°C for any zones of inhibition to develop. Results are summarized in Table 2.

The brine shrimp lethality results indicated that LC_{50} values for n-hexane (F1), dichloromethane (F3) and aqueous layer (F4) were 41.6 ppm, 31.3 ppm, 62.5 ppm and 62.5 ppm respectively - all of which are reasonably high activities.

Table 2 shows that the pure compound inhibited the growth of the Gram-positive bacteria namely *S. aureus, M. luteus, B. subtilis*, the Gram-negative bacterium, *E. coli* and *A. niger* - a fungus, with inhibition diameters of 2.6cm. 4.6cm, 2.6cm, 2.9cm and 1.6cm respectively.

The dichloromethane fraction (F3) and aqueous fraction (F4) showed reasonably high activities against *E. coli* and *S. aureus* but not on the other micro-organisms. The dichloromethane extract showed inhibition diameters of 3.6 cm and 4.3cm against *E. coli* and *S. aureus* respectively.

Aqueous fraction (F4) on the other hand showed inhibition diameters of 3.7 cm and 4.4 cm against *E. coli* and *S. aureus* respectively. The methanol extract did not show any activity when tested on the two strains of bacteria.

The results indicate that the pure compound, dichloromethane fraction (F3) and aqueous fraction (F4) contain active compounds that could be used to manage infections due to *A. niger, B. subtilis, M. luteus, S. aureus* and *E. coli.* Work is in progress to find out the active compounds in the fractions.

Table 1. Effect of *S. lyratus* fractures on brine shrimp

	% Deaths at 24 hours				
Fraction concentration	10ppm	25ppm	50ppm	100ppm	LC_{50}
Dichloromethane fraction	10	30	80	100	31.3
Methanol extract	10	10	40	90	62.5
Aqueous fraction (F4)	10	30	40	90	62.5

Table 2. Inhibition diameters (cm) at 24 hours of 100 ppm of samples

	Inhibition diameters (cm)				
	E. coli	*S. aureus*	*M. luteus*	*B. subtilis.*	*A. niger*
Dichloromethane fraction	3.6	4.3	Nil	Nil	Nil
Methanol extract	Nil	Nil	Nil	Nil	Nil
Aqueous fraction (F4)	3.7	4.4	Nil	Nil	Nil
Pure compound	2.9	2.6	4.6	2.6	1.6

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