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PRELIMINARY PHYTOCHEMICAL INVESTIGATIONS ON THE  
TWIGS OF CROTON MASTACHYUS - EUPHOBIAEAE FAMILY

BY

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A B S T R A C T

Investigations are going on on the East African *Croton* species to find out the active components in various parts of each plant. In this project, the petroleum ether extract of ground *Croton macrostachyus* twigs was found to yield crotepoxide as the major component. Thin layer chromatography showed presence of other compounds but these were in very minute quantities that could not be isolated. Work is going on to isolate some of these other compounds.

A summary of previous investigations on *Croton macrostachyus* is documented. In addition, a summary of all investigations on other *Croton* species carried out for the period 1983 - 1984 is given.

I N T R O D U C T I O N

*Croton* species - *Euphobiaceae* family are widely distributed in many parts of Africa. Many of them are used for medicinal purposes whereas others are poisonous. In Kenya, the following *Croton* species are found: *C. alienus*, *C. dichogamus*, *C. macrostachyus*, *C. megalocarpus*, *C. meryhartii*, *C. sylvaticus*, *C. pseudopulchellus* and *C. scheffleri* [1].

The leaves of *C. dichogamus* are dried and burnt by the Mbulu for inhalation by or fumigation of patients with fever and for chest ailments. The leaves are chewed or dried and smoked as cigarette by the Sukuma. It is also a remedy for stomach diseases. Chopped roots are added to soup from goats' meat and taken as a tonic. [2]

The seeds of *C. elliottianus* are used as a purgative. The oil from the kernel has the same effect but it is less irritant than the seed. The bark, mixed with curdled milk, is a Maasai purgative. Smaller doses of both the bark and the oil are diuretic. Both oil and seed are mildly anthelmintic but have no cholagogue action. The systematic action of seed and oil results in haemolysis and haemorrhagic spots in the tissues, the action of the oil being more feeble than that of the seed. [3]

*C. gratissimus* is used as a remedy for fevers. The Transvaal Sotho treat bleeding gums by brushing them with the charred and powdered bark. They also use the leaf as one of the ingredients for 'smoking' rheumatic patients. The plant is said to be very toxic and the Zulu use it as a cathartic and as an eruptive irritant. The bark is applied for its irritant action on the chest wall, in any painful respiratory condition, for intercostal neuralgia, for dropsy, indigestion and pleurisy. The Zulu use the powdered bark as one of the ingredients of a remedy inserted into the uterus for disorders of that organ. [3]

The bitter bark of *C. gubouga* is used in Transvaal and by Portuguese East Africans as a malaria remedy. In Gazaland and in Eastern Transvaal the bark has been used as a fish poison. Both the seed and the bark produce emesis and purgation in the dog and diarrhoea in the rabbit. In man they cause an intense burning sensation in the throat, salivation, nausea and slight purgation. These results are thought to be due to the presence of an acid principle. The Shangana take the seed as a drastic purgative. The bark contains a crystalline proline derivative, 4-hydroxyhygric acid,  $C_6H_{13}O_3N.H_2O$  mp.  $242^\circ$  which appears to be irritant and causes numbness on tasting. The Luvale administer an infusion of the root to thin babies to make them fat. [3]

The strongly scented roots of *C. jatrophoides* are used by Swahili people for colds and stomach aches. [2]

An infusion of the bark of *C. megalocarpus* is used by the Chagga, Luguru, Kamba and Kakamega people as an anthelmintic and for treating whooping cough. The oil from the seeds also has medicinal uses. [2]  
The Maasai use a decoction of the bark, with blood, as a tonic. [3]

*C. mubango* is used medicinally in Angola but no details are available [3] but Greshoff reports that it is a drastic purgative and vermifuge [4].

*C. menyhartii* root decoction is drunk by East Africans for treating influenza and malaria. [2]

*C. polytrichus* roots are used to treat headaches and labour pains by the Iraqw [2].

The boiled leaves of *C. pseudopulchellus* are applied to the chest for treating colds. Juice from boiled leaves and twigs is drunk for treatment of gonorrhoea. An infusion of the leaves is given to cattle as a remedy for anthrax. Leaves are burnt in among crops as an insecticide [2]. The Nyamwezi take a decoction of the root for

relief of asthma. The leaf and twig are used with the twig of *Teclea nobilis* in making a vapour bath for treatment of syphilitic sores, at the same time pounded root of *Crossopteryx febrifuga* being applied locally [3].

In Tanzania, *C. scheffleri* is used as a remedy for miscarriages [3].

A decoction from the root bark of *C. sylvaticus* is used by the Digo as a remedy for tuberculosis. An infusion of the leaves is taken as a purgative. A leaf decoction is used to wash the body for treatment of body swellings due to kwashiorkor or tuberculosis. Roots are pounded up to make poultices for swellings [2]. The bark is a Swazi remedy for gall sickness in cattle. The root is a remedy for pleurisy and indigestion and is said to contain the toxalbumin crotin. The bark is used in Gazaland as a fish poison. [3]

The roots of *C. zambesicus* are used as an aperient. The leaf decoction is used as a wash for fevers in Sierra Leone and S. Nigeria and internally for dysentery, fever and convulsions. The seeds are used medically in Togo [5]. The Maasai use the plant with *Grewia villosa* as an aphrodisiac [3].



Croton oil from the seeds of *C. tiglium* was formerly used in medicine as a very powerful purgative. The resin from the oil is more toxic to fish than rotenone. The seeds have been used in West Africa to poison human beings. [5]

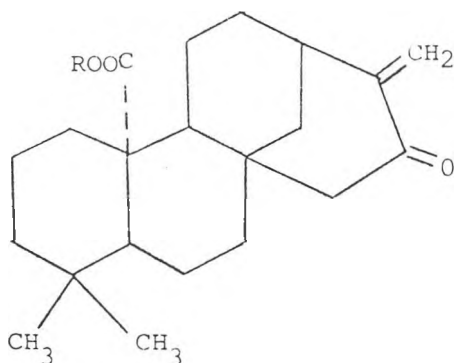
## CHAPTER ONE

WORK DOCUMENTED ON CROTON SPECIES FOR THE PERIOD 1983 - 1984

Although not many *Croton* species in Africa have been investigated pharmacologically and phytochemically, work on *Croton* species is on the increase in many parts of the world. Below is a summary of work done on *Croton* species over the period 1983 - 1984.

*Croton argyrophylloides*

The ethanol extract from *C. argyrophylloides* trunk wood afforded a new diterpenic acid (1a) and the corresponding methyl ester (1b), both with a  $\delta$ -lactonic ring system. Structural determinations were based on spectroscopic data and chemical reactions [6]

1a

R = H

1bR = CH<sub>3</sub>

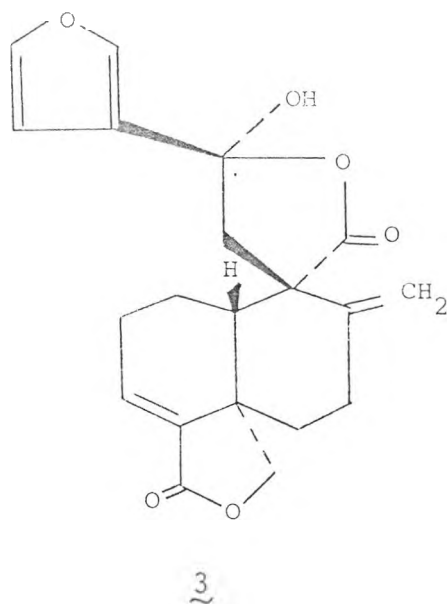


C. flavens

Hecker, E. *et al* have reviewed the epidemiology of *C. flavens* utilisation (especially as tea) by the natives of Curacao, West Indies and the concomittant high rates of oesophageal cancer among the population. [8]

C. jofra

The furanoid diterpene swassin (3) was isolated from dried stems of *C. jofra*. The structure of swassin was confirmed by  $^{13}\text{C}$ -NMR [9]

C. megalocarpus

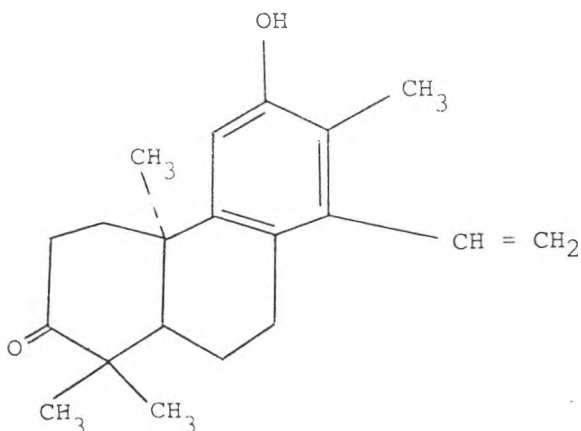
0-tetradecanoylphorbol - 13-acetate (TPA), 12-0-hexadecanoyl-16-hydrophorbol-13-acetate (HHPA), croton oil, tung oil and

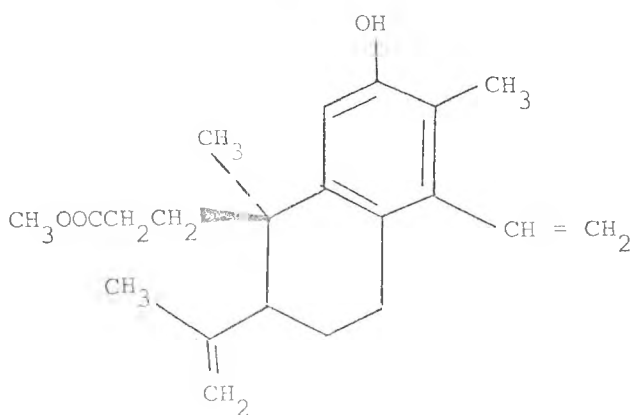
*C. megalocarpus* extract, known to possess Epstein-Barr virus

activating potency, were found to retain their capacity to induce Epstein-Barr virus early antigen complex in the viral genome - carrying lymphoblastoid cells even after heat treatment at 120°C for 2 hours or at 100°C for 12 hours. Such unusual heat resistance of the agents tested probably contributes to the persistence and accumulation of the active principle(s) in soil under the plants which contain such substance(s) [10].

C. sonderianus

Sonderianol (12-hydroxy-13-oxo gleistanth-8,11,13,15-tetraene) (4) and 3,4-secosonderianol (methyl-12,3-oate) (5), 2 new diterpenes with cleistanthane skeleton were isolated from heartwood of *C. sonderianus*. [11]



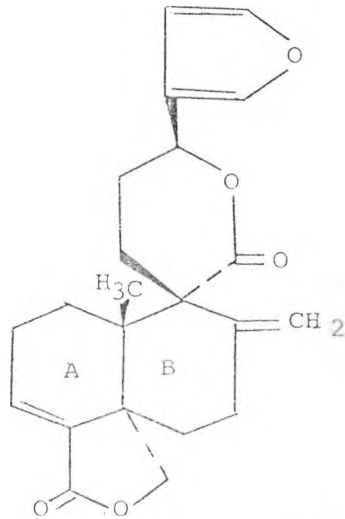


5

*C. sublyratus*

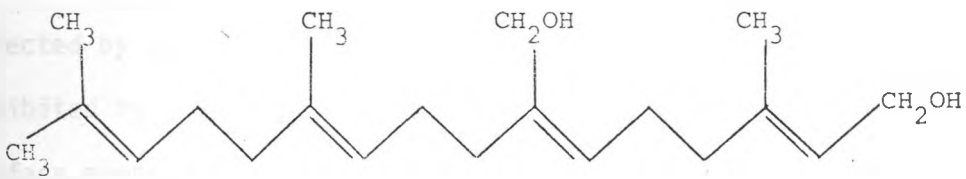
Extracts of *C. sublyratus* leaves gave 7 esters of 18-hydroxygeranyl geraniol: monoesters with stearic and oleic acid and 5 diesters (caprylic acid-palmitic acid, caprylic acid - oleic acid, 2 palmitic acid - oleic acid and linoleic acid-linolenic acid). Spectral and chromatographic characteristics of the 7 esters were established. The esters are known to have ulcer inhibiting activity [12].

X-ray crystallography of plaunolide (6) confirmed the chirality at C<sub>12</sub> and its distorted chair and skew-boat conformation of ring A and B respectively and that both the  $\gamma$ -lactones are fused to the octalin ring in envelope conformations [13].



6

A diterpene alcohol (7) was prepared by extraction of *C. sublyratus*, a plant used in ulcer treatment followed by the resultant extracts with bases. 2.8 kg of crushed *C. sublyratus* was refluxed with 10 l methanol, concentrated to 5 l, refluxed with 1 l 10% aqueous NaOH for  $\frac{1}{2}$  hour and purified with silica gel chromatography in ethyl acetate-benzene to give 2.04 g of (7) [14].



7

### *C. tiglium*

The kinetics of haemolysis of rabbit erythrocytes by *C. tiglium* lectin was studied as a function of concentration of the lectin and erythrocytes. The length of the prelytic period decreased with

increased lectin concentrations, indicating that the secondary events at the membrane which follow the binding of the lectin to cell surface carbohydrate receptors are accelerated at higher surface concentrations of the lectin. The rate or extent of haemolysis was not affected by the inclusion of ions like  $K^+$ ,  $Ca^{2+}$ ,  $Mg^{2+}$  in the medium or by the substitution of ionic medium by non-ionic medium. The inhibition of haemagglutination and haemolysis of rabbit erythrocytes by *C. tiglium* lectin by antilectin rabbit serum was observed [15].

*C. tiglium* lectin (CTL), a protein with haemagglutinating and haemolytic activities which is specific for only complex carbohydrates, agglutinates phospholipid-glycolipid vesicles in the presence of 1 mM  $CaCl_2$ . The CTL-induced agglutination of liposomes, which is not affected by phospholipid compounds and ionic strength, is completely inhibited by trypsin-released glycopeptides from sheep erythrocyte surface membranes, indicating that the phenomenon is mediated by lectin - carbohydrate interactions. Since the lectin reactive glycolipids all carry the sequence Galactose - Galactose on their N-acetylated derivatives as the common structure denominator, it appears that the disaccharide unit Galactose-Galactose or their N-acetylated derivatives constitute an essential part of the



haptens of the lectin. Lack of evidence for non-carbohydrate dependent hydrophobic interaction of CTL with phospholipids and glycolipids lends support to the view that haemolysis is also a carbohydrate-dependent function of the lectin [16].

#### Croton species

Polysaccharides were prepared from gingseng root, Mows root bark, phellodendron bark, *caesalpinia sappan* and croton seeds. An aqueous extract of the mixed polysaccharides from the latter three species were also prepared. The gingseng polysaccharides and the mixed aqueous extract had antitumour activity against Sarcoma 180 in mice. Studies with various cellular components of the immune system suggested that the mixed aqueous extract stimulated the tumour related immune system with no effects on the overall immune system, whereas the gingseng polysaccharides appeared to stimulate the latter. [17].

CHAPTER TWOPREVIOUS WORK DONE ON *CROTON MACROSTACHYUS*

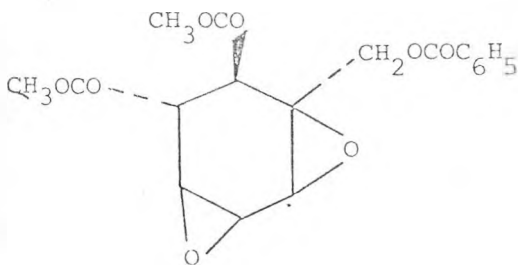
*Croton macrostachyus* is a tree upto 50 feet high with white flowers and slightly 3-lobed fruits  $\frac{1}{2}$  inch in diameter. The habitat is the savannah forest. It is found in many parts of Tropical Africa; from Guinea to Cameroon in West Africa to parts of East and Central Africa including Kenya. [5]

The Chagga use *C. macrostachyus* and the leaves of *Embelia kilimandscharia* as an anthelmintic. In Ethiopia it is used as a taenifuge [3]. In East Africa leaves are boiled and the decoction drunk for coughs. Root decoction is used as an anthelmintic for tapeworms and as a purgative. Ash from burnt leaves is licked for coughs. Juice from a fresh leaf is applied on fresh wounds to hasten blood clotting. Juice from boiled roots is drunk for malaria and venereal diseases. The bark is peeled from stems and roots boiled in water and the newly born baby bathed in the mixture as a remedy against skin rashes. The seeds and resin are however, poisonous. [2]

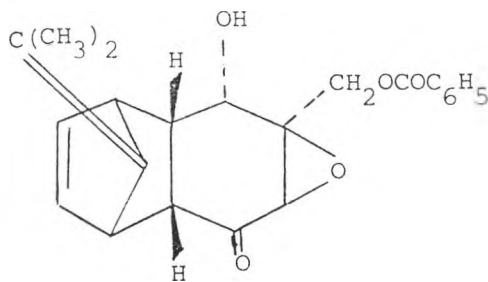
Up till 1984 only the following work had been done on *C. macrostachyus*:

A short term in vitro assay for promoter substances using human lymphoblastoid cells latently injected with Epstein-Barr virus (used to assay such substances from *Euphobiaceae* family including *C. macrostachyus* and *C. megalocarpus*). [18]

The structure of dl crotepoxide (8) previously isolated from the seeds of *C. macrostachyus* and *Piper futokadzura* was confirmed by synthesis from epoxide (9) in 9 steps. [19]

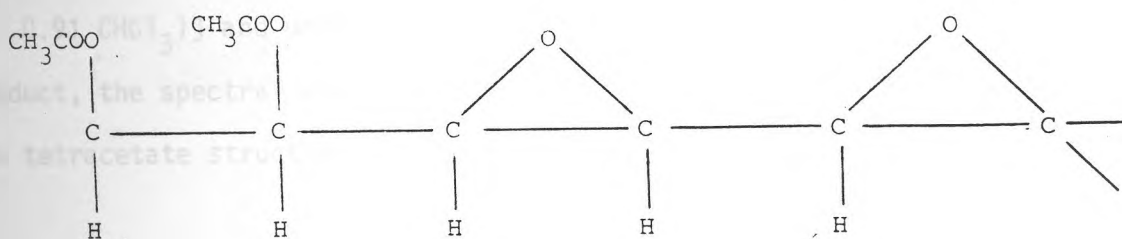


8



9

Fractionation of the ethanol extract of the fruits of *C. macrostachyus* [20], guided by assay against Lewis Lung carcinoma, revealed that an active principle was concentrated successively in the methanol layer of a 10% aqueous Methanol-Skellysolve B partition and in the 1-butanol layer of a 1-butanol-water partition. Further fractionation involving silicic acid chromatography yielded crotepoixide,  $C_{18}H_{18}O_8$  mp 150 - 151°,  $[\alpha]_D^{25} + 74^\circ (c 1.70 \text{ CHCl}_3)$ ;  $\lambda_{\text{max}}^{\text{MeOH}}$  274 m $\mu$  ( $\epsilon$  1050) and 281 m $\mu$  ( $\epsilon$  860);  $\lambda_{\text{max}}^{\text{CHCl}_3}$  3.35, 5.71, 5.78, 6.24, 6.31, 6.89, 7.29, 7.87, 8.20, 9.00, 9.60, 10.24 and 11.12 $\mu$ . NMR signals (in  $\text{CDCl}_3$ ) at  $\tau$  2.28 (5H, m, aromatic); 4.27 (1H, d,  $J_{xy} = 9.5$  Cps > CHOAc); 5.02 (1H, d, d,  $J_{xy} = 9.5$  and  $J_{AY} = 1.5$  cps > CHOAc); 5.42 and 5.75 (2H, doublets,  $J = 12.0$  cps,  $\text{CH}_2\text{OCOPh}$ ); 6.32 (1H, d,  $J_{BC} = 2.5$  cps); 6.56 (1H, d, d,  $J_{AB} = 4.0$  and  $J_{AY} = 1.5$  cps); 7.88 (3H, s, acetate) and 7.95 (3H, s, acetate).



Crotopoxide was converted to several crystalline derivatives.

Hydrogenation using platinum oxide catalyst yielded the hexahydro-  
derivative  $[C_{18}H_{24}O_8]$ ; mp 121 - 122°;  $[\alpha]_D^{30} + 59$  (C1.35,  $CHCl_3$ )

which exhibited no signals for the aromatic protons in the NMR  
spectrum. Treatment with aqueous methanolic potassium hydroxide

yielded a triol (10)  $[C_7H_{10}O_5]$ ; mp 101 - 102°;  $[\alpha]_D^{27} + 30^\circ$   
(C.106  $CH_3OH$ ) and benzoic acid. Treatment with aqueous methanolic

HCl for 30 minutes yielded the monochlorohydrin (11)  $[C_{18}H_{19}ClO_8]$ ;  
mp 170 - 171°;  $[\alpha]_D^{29} - 4^\circ$  (C 1.35,  $CHCl_3$ ) while prolonged

treatment yielded the deacetyl dichlorohydrin (12)  $[C_{14}H_{16}Cl_2O_6]$ ; mp  
241 - 242°;  $[\alpha]_D^{25} - 10^\circ$  (C 0.71,  $CH_3OH$ ). (12) was readily

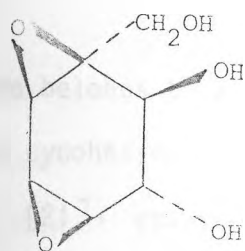
converted to a triacetate  $C_{20}H_{22}Cl_2O_9$  mp 217 - 218°; and under  
more drastic conditions to tetracetate  $C_{22}H_{24}Cl_2O_{10}$  mp 153 - 154°.

Treatment of crotopoxide with aqueous methanolic HI yielded a  
monoiodohydrin (13)  $[C_{18}H_{19}IO_8]$ ; mp 143 - 144°  $[\alpha]_D^{28} - 46^\circ$

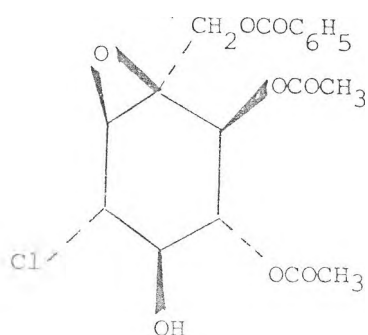
(C.1.4  $CHCl_3$ ) and an ene diol derivative (14)  $[C_{18}H_{20}O_8]$ ; mp  
145 - 146°;  $[\alpha]_D^{28} + 127$  (C. 1.51  $CHCl_3$ ). The ene diol was

converted into triacetate  $[C_{20}H_{22}O_9]$ ; mp 141 - 142°  $[\alpha]_D^{28} + 151$   
(C. 0.91  $CHCl_3$ ) and under more drastic conditions into an oily

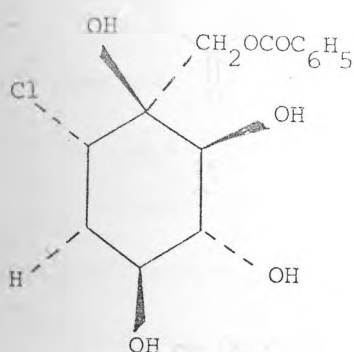
product, the spectral characteristics of which are in agreement with  
the tetracetate structure.



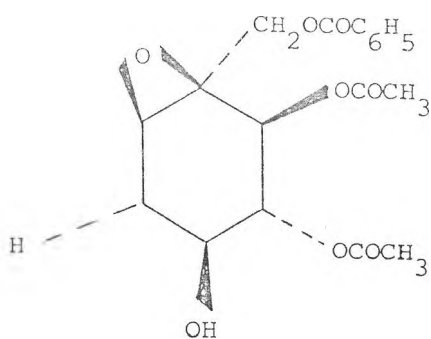
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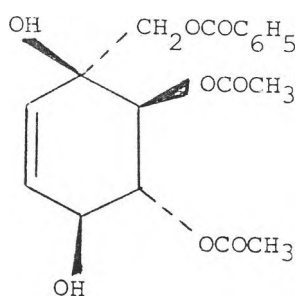
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12



13



14

Crotopoxide has been found to possess tumour inhibitory activity in Lewis Lung carcinoma in mice at 200 mg/kg and Walker carcinosarcoma - 256 in rats at 300 mg/kg.

Although *Croton* species are widely used throughout Africa for medicinal purposes, very little is known about the active components in these plants. Most of the studies so far reported on crotons have been on species occurring in South America, West Indies and a few of the West African species. East African species have received virtually no attention. In the case of *Croton macrostachyus* only the fruits have been investigated. This project is a part of an on going programme of phytochemical investigations on East African crotons in order to find out the chemistry of the active components in them. Since different parts of these plants have been used for different medicinal purposes, the active components in different parts of the plant need to be identified. In this project, phytochemical investigations are carried out on the twigs of *Croton macrostachyus* with the aim of isolating the main component(s) in a pure form and characterising it (them).

It also absorbed at UV  $\lambda_{\text{max}}^{\text{MeOH}}$  235 nm and 279 nm (figure 2) which is very similar to the UV absorption for crotepoixide which is at  $\lambda_{\text{max}}^{\text{MeOH}}$  231 nm and 276 nm (figure 3). This faster-moving compound is therefore probably in the same class of compounds as crotepoixide, that is, an epoxy-cyclohexane derivative, but probably less polar or of lower molecular weight. Work on this compound is still going on to determine its structure.

The Rf values of the major constituent, that is, the white needle-like crystals were found in different solvents (figure 4) to be as follows:-

Solvent	Rf values
Benzene : chloroform = 1 : 1	0.215
Benzene: chloroform = 1 : 3	0.310
Toluene: Ethyl acetate = 9 : 2	0.750
Diethylether : chloroform = 1 : 3	0.103
Benzene	0.219

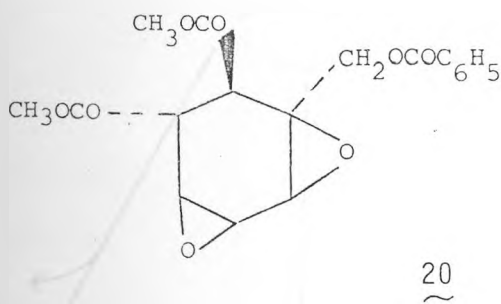
Its m.p., which was 145 - 146°C on Gallenkamp mp apparatus and 147 - 149°C on Koffler Hot-stage mp apparatus, and its optical rotation,  $[\alpha]_D^{20} + 70$ , are very similar to those reported for crotepoixide, first isolated



from the seeds of *C. macrostachyus* (mp - 151° and  $[\alpha]_D^{25} + 74$ ).

IR (KBr) absorption (figure 5) showed peaks at 3100 - 2900  $\text{cm}^{-1}$  (C - H stretching of  $-\text{CH}_2-$  and  $-\text{CH}_3$ ); 1765  $\text{cm}^{-1}$  (aliphatic ester carbonyl); 1725  $\text{cm}^{-1}$  (benzyl ester carbonyl); 1600, 1580, 1450  $\text{cm}^{-1}$  (aromatic C = C stretching); 1430, 1370  $\text{cm}^{-1}$  (C - H bending of  $-\text{CH}_3$ ); 1280, 1230, 1210, 1180  $\text{cm}^{-1}$  (C - C of benzene and epoxy C - H).

The  $^1\text{H}$  and  $^{13}\text{C}$  - NMR spectra (figures 6 and 7 respectively) unambiguously established the compound as crotepoxide (20) (see experimental for details).



Due to the facile loss of  $\text{C}_6\text{H}_5\text{C}=\text{O}$ , no  $\text{M}^+$  could be seen in the EIMS (figure 8). However, the fragmentation pattern, coupled with the evidence from the  $^1\text{H}$  and  $^{13}\text{C}$  - NMR were consistent with the compound being crotepoxide.



The other components detected ( $\text{GN}_2$ ,  $\text{GN}_3$  and  $\text{GN}_4$ ), gave various ranges of colour with anisaldehyde reagent, suggesting they might be terpenoidal in nature. Apart from their  $R_f$  values, which were measured in chloroform; benzene = 1:1, no further work was done on them.

CHAPTER FOUREXPERIMENTAL

Analytical TLC was carried out with silica gel 60 GF<sub>254</sub> as the adsorbent. The plates were prepared by conventional methods. Column chromatography was on silica gel 60 (0.063 - 0.200 mm  $\approx$  70 - 230 mesh ASTM).

IR spectrum was recorded on a Perkin-Elmer IR spectrophotometer 727B and UV spectrum on a Pye Unicam sp 8000 UV spectrophotometer.

$[\alpha]_D$  was determined in chloroform on Perkin Elmer digital polarimeter.

Mass spectrum was determined on a Finnigan 4000 and on a MAT 312/SS 200 instrument with Ei at 70 eV.

<sup>1</sup>H and <sup>13</sup>C - NMR spectra were determined at 90 and 25 MHz respectively on a Jeol FX 90 Q instrument, with TMS as the internal reference and CDCl<sub>3</sub> as the solvent.

Melting point was determined using Gallenkamp mp apparatus as well as Koffler hot-stage melting point apparatus.

Anisaldehyde spray reagent (1% w/v anisaldehyde: 1% v/v concentrated

sulphuric acid in glacial acetic acid) was used for visualisation of TLC spots.

The plant material was collected from Karatina, about 120km north of Nairobi in July/August 1985.

#### Extraction

590g of *A* ground twigs of *Croton macrostachyus* were extracted with about 3½ l petroleum ether (bp 60 - 80°) in a Soxhlet apparatus for 48 hours. The extract was reduced to about 150 mls on a rotary evaporator, and stored in the refrigerator for 5 days during which a brown solid was deposited. The crude extract was filtered to yield 2.06 g of a brown residue. The filtrate was concentrated to about 40 mls and stored in the refrigerator. A small amount of the residue obtained was dissolved in chloroform and TLC was done on it using microscope slides to find the best solvent systems. These were found to be chloroform, in which it moved faster and benzene, in which it moved slowly. The rest of the residue was dissolved in minimum amount of chloroform (4.2 mls) and column chromatography was carried out on it using 1 liter benzene, 750 mls benzene; chloroform = 5 : 1, 1 liter benzene : chloroform = 1 : 1, 1 liter chloroform and 500 mls chloroform: methanol = 1:1 in the order given. 15 ml fractions were collected and they were monitored by TLC. Samples of similar

Composition were combined and labelled as follows:-

GN<sub>1</sub> was samples 191 - 290 and was a pale yellow solution

GN<sub>2</sub> was samples 140 - 190 and was a colourless solution

GN<sub>3</sub> was samples 1 - 139 and was colourless

GN<sub>4</sub> was all the remaining eluate and it was a brown solution.

#### Work done on GN<sub>2</sub> and GN<sub>3</sub>

GN<sub>2</sub> and GN<sub>3</sub> were each evaporated to dryness and dissolved in minimum amount of methanol. After staying in the refrigerator for a few days each solution deposited a solid. On filtration both GN<sub>2</sub> and GN<sub>3</sub> each produced a cream-grey greasy substance in very small amounts and no further work was done on the solids. The mother liquor from GN<sub>2</sub> showed components with Rf 0.333 and 0.235 and that of GN<sub>3</sub> six components with Rf 0.204, 0.322, 0.500, 0.605, 0.737 and 0.875 both in chloroform : benzene = 1:1 (figure 9). No further work was done on them.

#### Work done on GN<sub>4</sub>

GN<sub>4</sub> was evaporated to dryness and dissolved in minimal amount of methanol. After storage in the refrigerator for some days GN<sub>4</sub> deposited a brown solid (15.8 mg) which showed two components with Rf 0.061 and 0.959 (figure 9) while its mother liquor showed three components with Rf 0.299, 0.240 and 0.175 (figure 9) both in chloroform : benzene 1:1.

Detection of these components was with anisaldehyde reagent in which they gave various colours, indicating they might be terpenoids.

#### Work done on GN<sub>1</sub>

GN<sub>1</sub> was evaporated to dryness and dissolved in minimal amount of methanol. After staying overnight in the refrigerator, the solution yielded white crystals. It was filtered and the crystals were recrystallised from methanol to give 91.0 mg of white needle-like crystals. The mother liquor of GN<sub>1</sub> was found to contain one other compound in very small quantities which did not recrystallise. The compound was faster running than GN<sub>1</sub> crystals.

#### Work done on GN<sub>1</sub> crystals

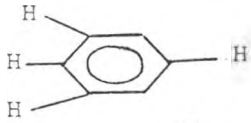
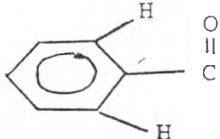
m.p. 145 - 146° on Gallenkamp mp apparatus and 147 - 149°C on Koffler hot-stage mp apparatus.

Optical rotation using 0.63 mg/ml solution in chloroform:  $[\alpha]_D^{20} + 70^\circ$

The compound absorbed at UV  $\lambda_{\max}$  231 nm  $\epsilon = 9,115$  and  $\lambda_{\max}$  276 nm  $\epsilon = 1,089$  in 0.028 mg/ml methanol (Literature values:  $\lambda_{\max}^{\text{MeOH}}$  274 nm  $\epsilon = 1,050$  and 281 nm  $\epsilon = 860$  [20]).

IR spectroscopy (KBr) gave peaks at 3100 - 2900, 1765, 1725, 1600, 1580, 1450, 1430, 1370, 1320, 1280, 1230, 1210, 1180, 1120, 1070, 1040, 1010, 910, 900, 860, 710  $\text{cm}^{-1}$ .

$^1\text{H}$  - NMR signals in  $\text{CDCl}_3$  with TMS as the reference:

<u><math>\delta</math></u>					
2.025	s	3H			$\text{CH}_3\text{CO-}$
2.119	s	3H			$\text{CH}_3\text{CO-}$
3.1	d,d	1H	$J \sim 4.2, J_2 \sim 2$	}	3 Epoxy protons
3.4	t	1H	$J \sim 4.2$		
3.65	d	1H	$J \sim 2.8$		
4.15, 4.29	d	1H	$J \sim 11.3$		$-\text{CH}_2\text{OCOC}_6\text{H}_5$
4.51, 4.65	d	1H	$J \sim 11.3$		
4.9	d,d	1H	$J \sim 9.8$		$\text{>CHOCO}$
5.7	d	1H	$J \sim 8.4$		$\text{>CHOCO}$
7.5	m	3H			
8.0	m	2H			



$^{13}\text{C}$  - NMR signals in  $\text{CDCl}_2$

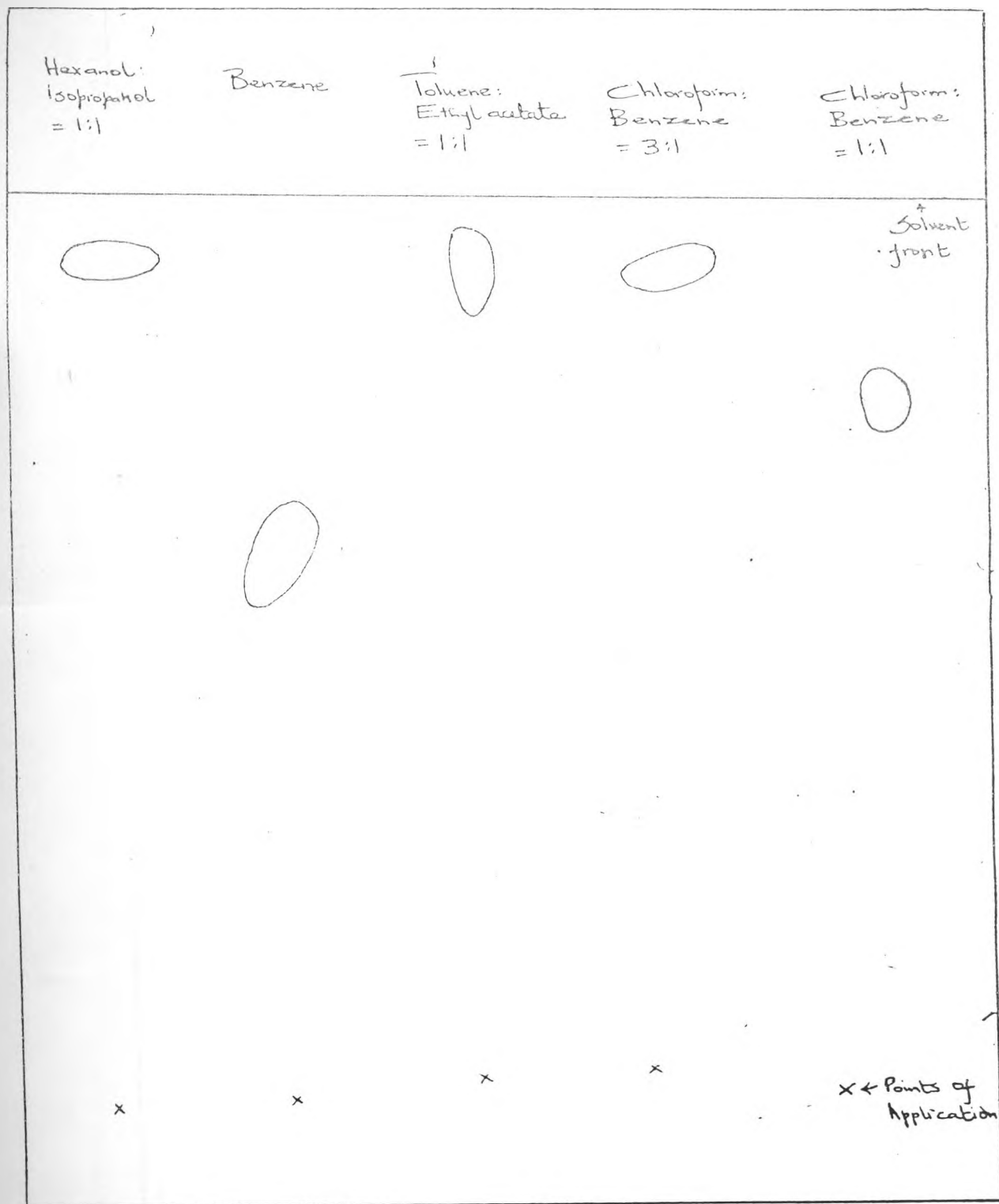
$\delta$  values

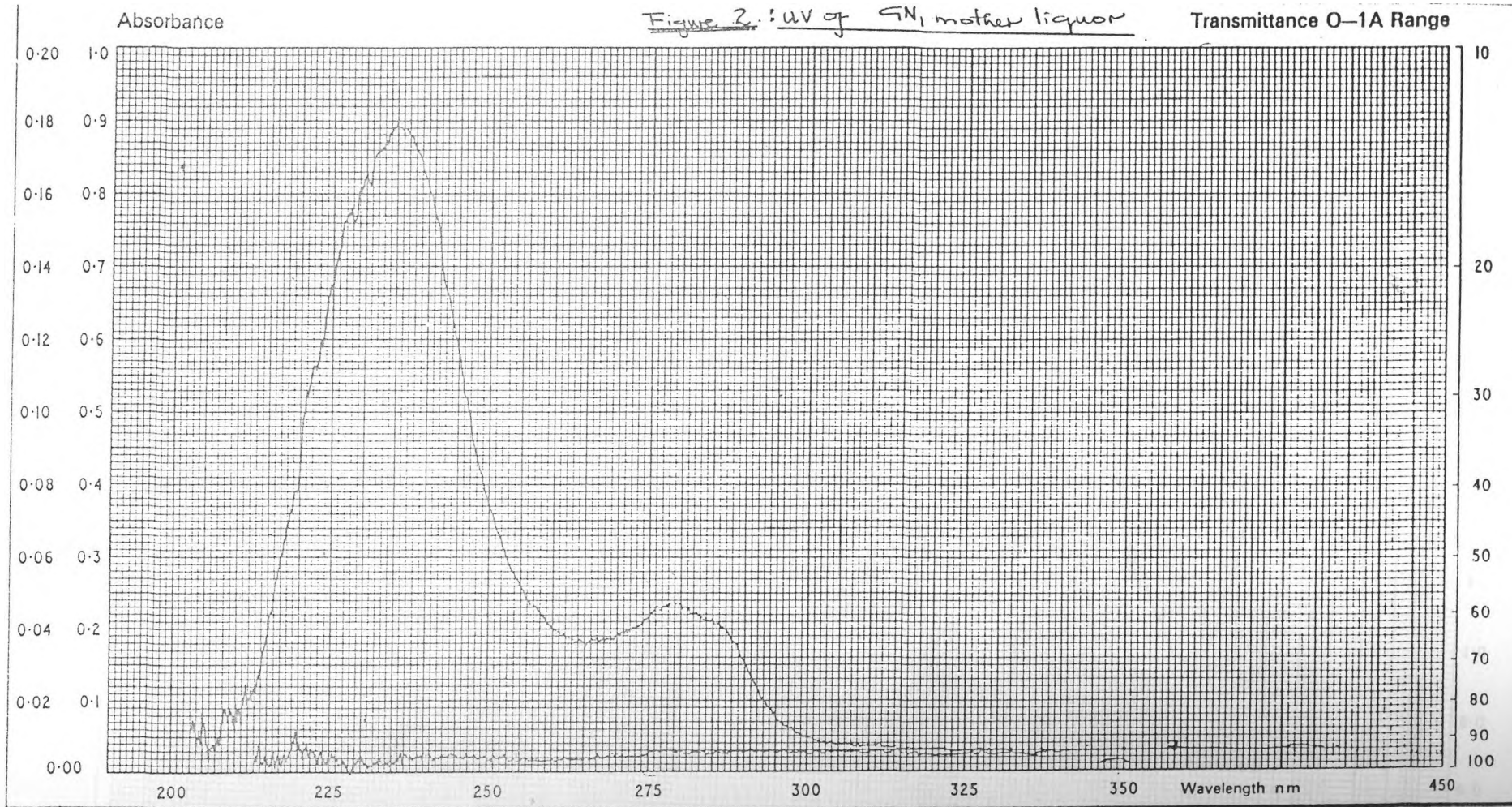
20.415	}	$\text{CH}_3\overset{\text{O}}{\parallel}{\text{C}}-\text{O}-$
47.935		$\text{CH}_2-\text{O}-\overset{\text{O}}{\parallel}{\text{C}}-$
52.486	}	cyclohexane carbons
53.623		
59.340		
62.400		
69.551		
70.336		
128.411	}	Benzene carbons
129.197		
129.657		
133.314		
165.602	}	$\overset{\text{O}}{\parallel}{\text{C}} - \text{Ar}$
169.475		}
169.800		

Mass spectrum showed prominent peaks at  $m/z$  43, 77, 97, 105, 115, 163, 207, 227, 232, 249. (figure 8).

Figure 1

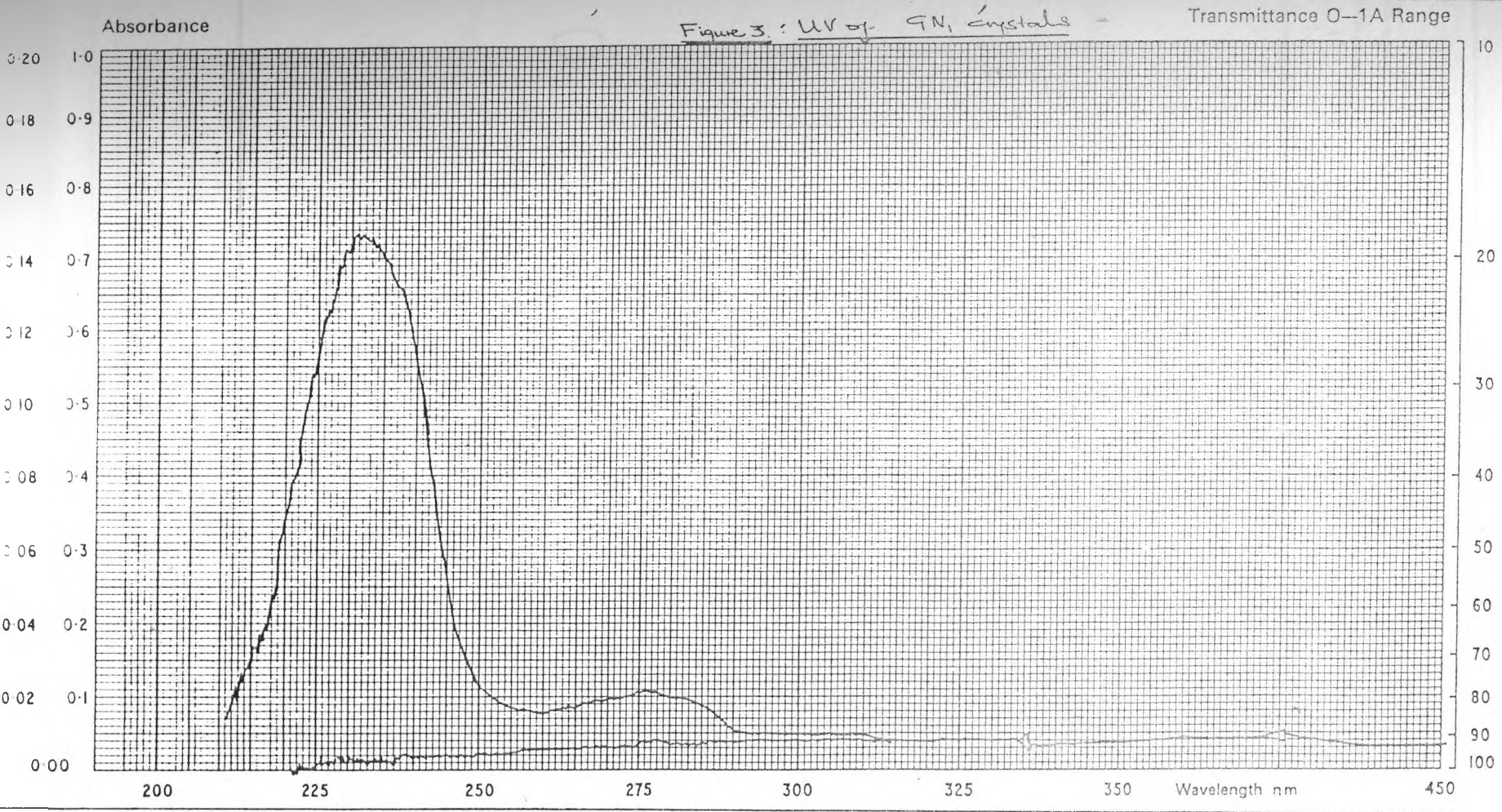
Chromatograms of the mother liquor of 9H<sub>1</sub>





Reference: Methanol.

Figure 3: UV of  $\text{qN}_1$  crystals



Reference: Methanol.

Figure 4

Chromatograms of  $\text{G.N.}$  crystals in various solvents

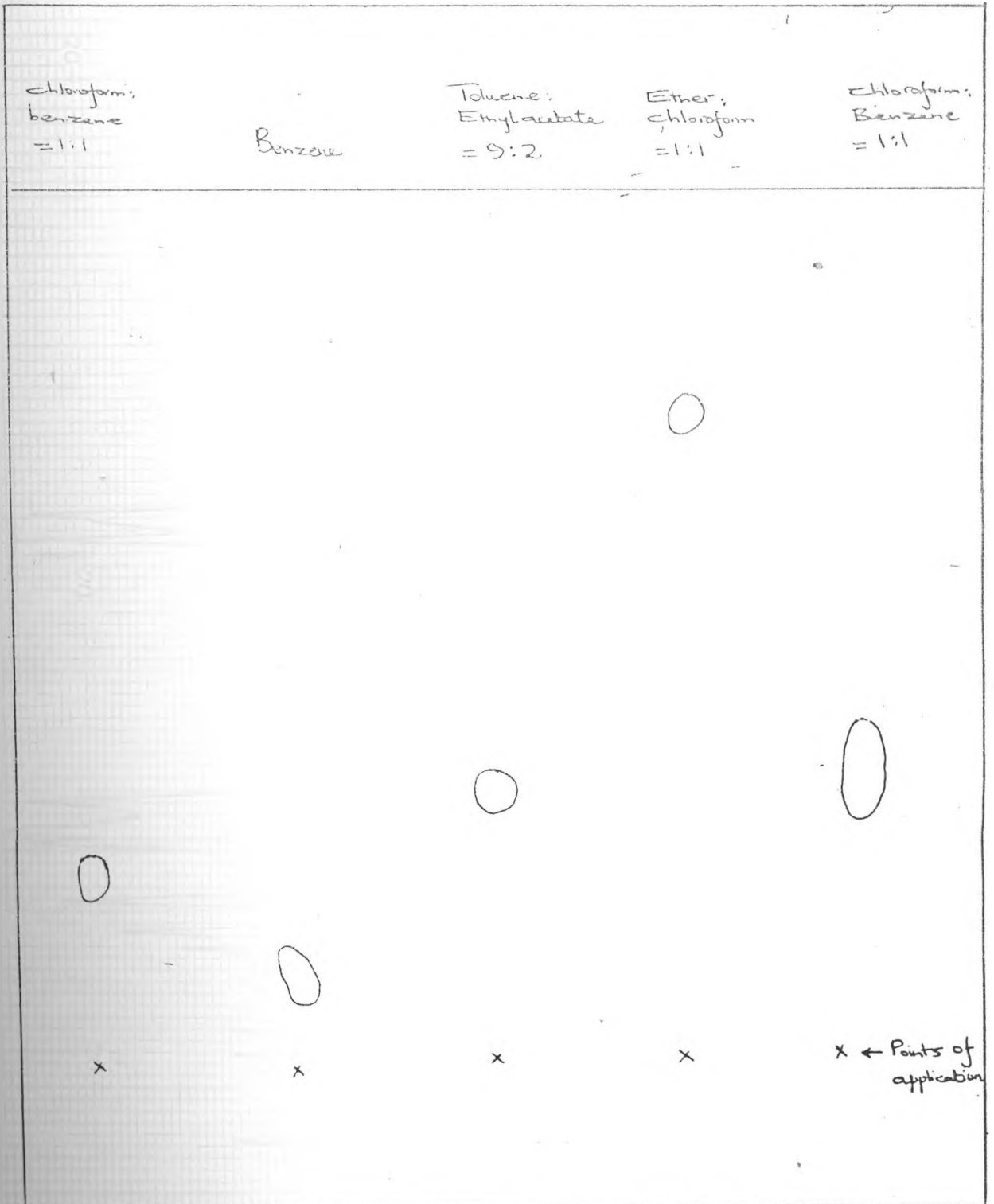
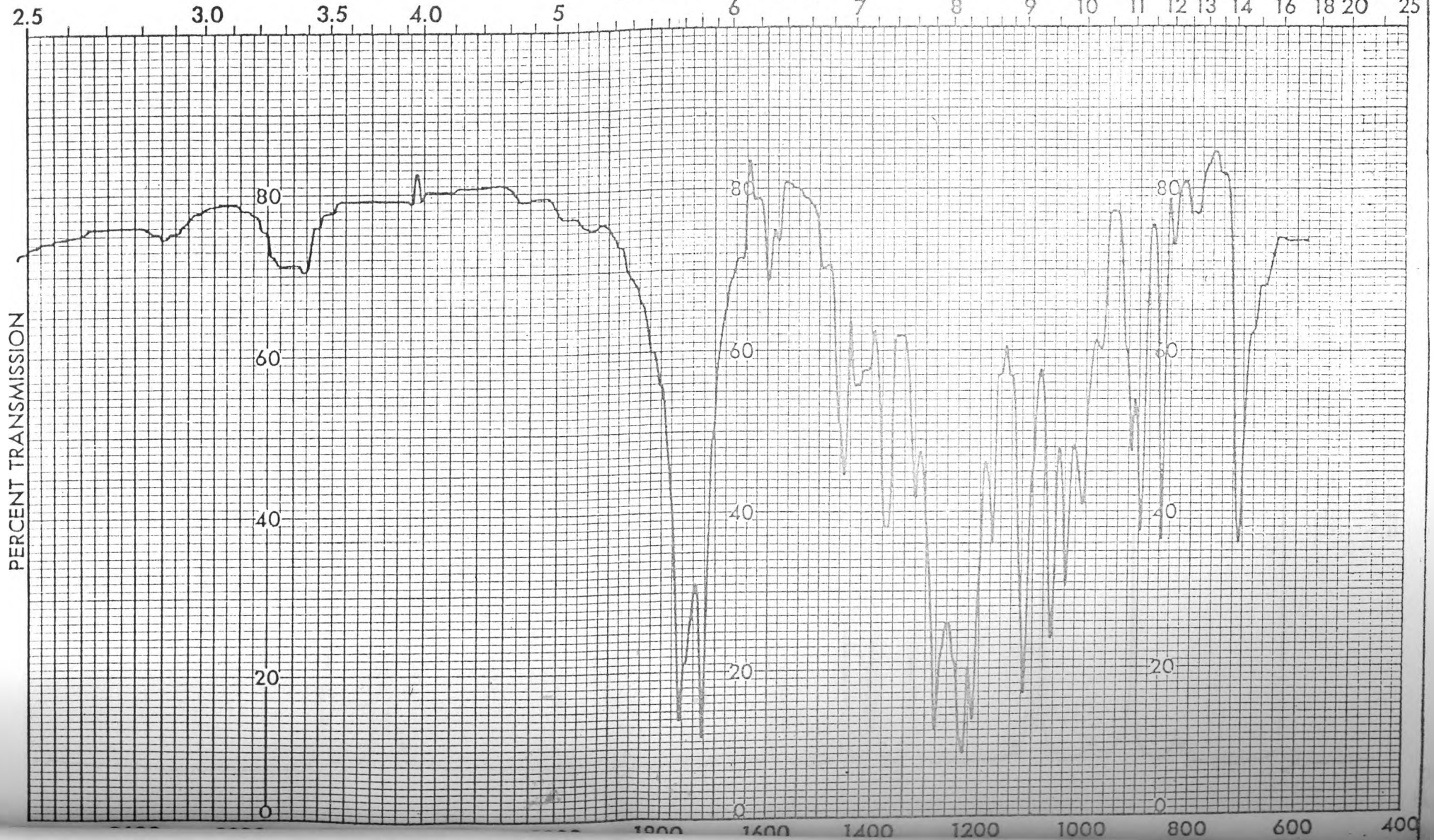


Figure 5 : IR of  $\text{Si}_3\text{N}_4$  crystals.





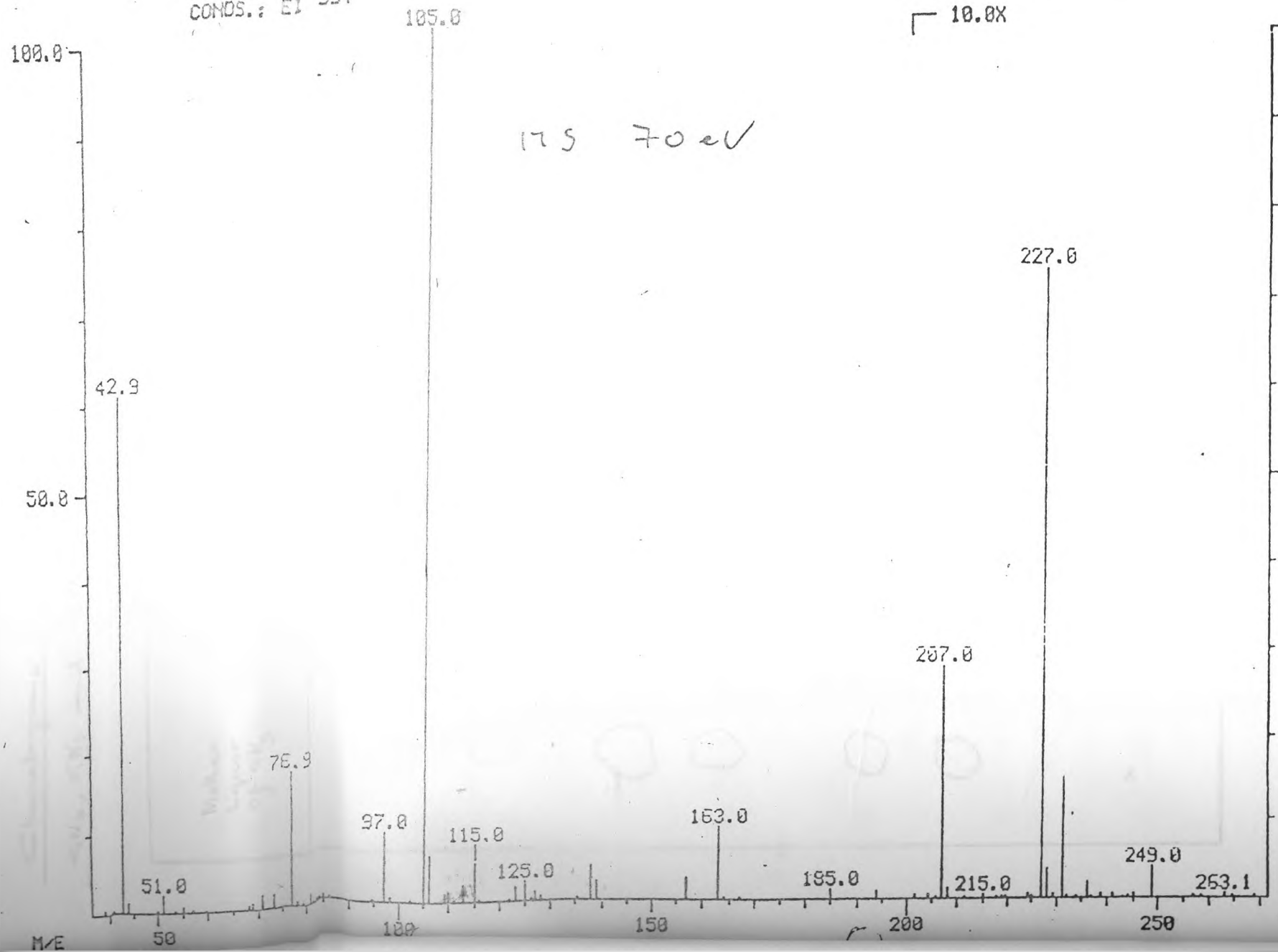




SAMPLE: GM1  
CONDOS.: EI SST

BRSE M/E 105  
10.0X

40



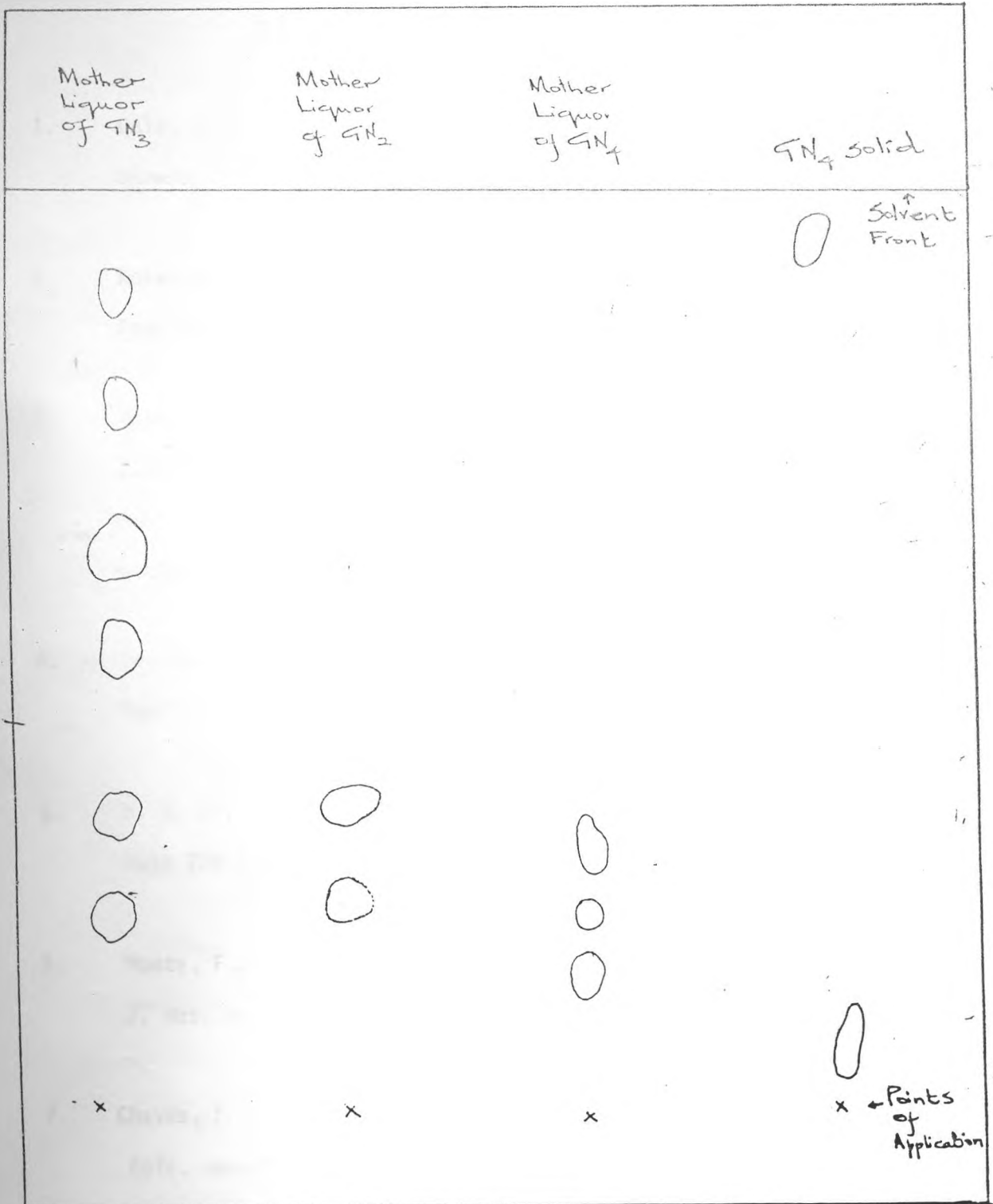
175 70 eV

42

17344.

Figure 9

Chromatograms of  $GN_1$  and the mother liquors of  $GN_2$ ,  $GN_3$  and  $GN_4$



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