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IDENTIFICATION OF RAW TECHNICAL METALAXYL STRUCTURE AND THE MAIN IMPURITIES ASSOCIATED WITH IT.

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ABSTRACT

A new analytical method was devised using gas chromatography with tanderm mass spectrometry (GC-MS) to identify and confirm metalaxyl and its impurity. Their compounds show similar fragmentation patterns indicating numerous single bond. All compounds have unique retention time on a SE-30 capillary column and common ions form most of the parent fungicide and its impurity allow multiple-ion monitoring for confirmation. This study consider a basic analysis in pest. Analysis Res. department.

INTRODUCTION

Methyl [N-2,6-dimethylphenyl]-N-(methoxyacetyl)–DL-laninate (see structure I in fig. 1) is the active ingredient of the fungicide ridomil. Since its discovery in 1977, it has been widely used for the control of plant diseases caused by 60 mycetous fungi of the order peronosporales *(Schwinn et al., 1977; Houseworth, 1987)*. It has low mammalian toxicity and regards as safe for general use. However combination of trace impurities present in technical material (either during synthesis or storage), may lead markedly to different more toxicities than would be expected for the toxicities of the individual components, (*Pelligrini and Santi ;1972)*.

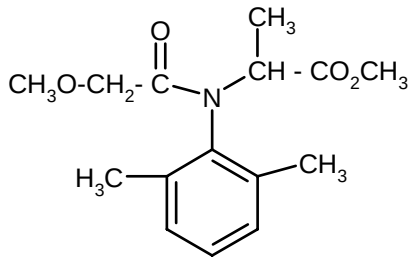
This study was undertaken to obtain gas chromatographic mass spectra of the parent Metalaxyl and to elucidate fragmentation patterns so that subsequent residue studies could be conducted with MS confirmation or single or multiple ion quantization. Also this investigation is concerned with the identification of the impurities,

which are commonly present or may be developed upon storage of technical metalaxyl.

MATERIALS AND METHODS

1. Pesticide test:

The structure of technical fungicide metalaxyl 98% is as follow:



IUPAC name: methyl N-(methoxyacetyl)-N-(2,6-xylyl)-DL-alaninate; methyl 2-{[(2,6-dimethylphenyl)methoxyacetyl]amino}propionate

1.A Active ingredient

1.A.1. Metalaxyl as mentioned by CIPAC E, 365/TC/M/3 (1993).

1.B. Impurities

1.B.1. 2,6-dimethylaniline as reported by CIPACE, (365/TC/M/4, 1993).

Maximum: 1g/kg: as indicated by FAO **(1995).**

Experimental section:

1. GC Conditions:

Gas chromatography was performed on a Hewlett Packard Model 6890 instrument equipped with a flame ionization detector (FID). A fused silica capillary column (10m length, 0.53 mm-id, coated with methyl silicon gum HP-1, with 2.65 μm film thickness) was used. Nitrogen was used as carrier gas with a flow rate of 35 ml/min (Metalaxyl) or 30 ml/min (2,6-dimethy/aniline). The injector (split less capillary inlet system, injection volume 1 μL and detector temperature 260°C (metalaxyl) or 290°C (2,6-dimethy/aniline respectively. Oven temperature was programmed 205°C (metalaxyl) or 75°C for 2 minutes then 75°C to 90°C with 6°C/min, then 90 to 260°C with 15°C / minute then 290°C for 5 minute (2,6 di methyl

aniline) for the determination of Metalaxyl and its impurities.(Ripley and Braun 1983); Sukul et al. ,1992 and CIPAC E.; 1993).

2. Gas chromatography – Mass spectrometry (GC-MS):

GC-MS was conducted an Agilent 6890 mass selective detector (MSD). An Agilent 6890 gas chromatograph with a 15 m x 0.25 mm fused silica capillary column with 0.25-μm coating of SE-54 and He carrier gas flow of 2ml min^{-1} was connected to the MSD with a direct capillary interface operated at 260°C. one-micro liter solutions were injected with a split less injector operated at 220-250°C with the bypass valve open for 1min. The column oven was programmed for an initial 1-min hold at 90°C, followed by a 10°C min^{-1} rise to 270°C and a final hold at the temperature to allow elution of late compounds.

The MSD was optimized by using Agilent disk software under Auto tune conditions PFTBA (PerFluoro Tributylamine) calibration. Mass spectra were acquired over the 40-400 amu range at 380 amu s^{-1} and normalized to DFTPP (Deca Fluoro Triphenylphosphine).

Single-or multiple ion monitoring was conducted with Agilent software with up to 20 ions being monitored.

N.B.: The mass spectrometric detector (MSD) was operated in electron impact ionization mode, scanning from m/z 50 to 500. The ion source temperature was 220°C and the quadruple temperature 150°C. The electron multiplier voltages (EM voltage) was maintained 1100v above autotun, and solvent delay of 3 min was employed.

RESULTS AND DISCUSSION

Part 1. Metalaxyl

A. Metalaxyl content retention time and data were generated on a GC as follow: Retention time = 4.5 min. and estimation active ingredient in Metalaxyl Tech. = 97.7%

B) Mass spectra.

The mass spectra of metalaxyl (Marucchini et al. 1983; Ripley and Braun 1983 and Cooke et al., 1982) have been reported and tentative fragmentation patterns have been made (Cooke et al. 1982; Ripley 1985). This study confirms that the examined metalaxyl follow this general pattern as shown in Fig. 2. The metalaxyl compound exhibits weak to moderately intense molecular ions and numerous characteristic ions at moderate intensity were found at even m/z values

indicating single bond cleavage. The base peak in most cases arises from cleavage of d-e or f-g (Figure 2). The base peak (M-73) is a major fragment ion and loss of neutral CO from the substitutedacyl fragment or d-e cleavage on the parent molecule results in a base peak of m/z 45; [m-30] ([M-OCH_3] + H) and [M-45] are also evident. This fragmentation tends to predominate regardless of other substations as its shown Table (1).

Cleavage of K-I [M-59] or loss of $COOCH_3$ from other ions predominates. The R_1 alkyl substituent on the aniline tends to stabilize the fragment and m/z 132 is common to most of the spectra. Most of the ions for the other simple fragmentations from the molecular or fragment ions are present in the mass spectrum. Metalaxyl shows ions for M-30; -45; -59, -73, -87,-105, -117, -119, -131, 133 and 147. A number of other degraded products having various R_1 and/or R_2 similar to those of the parent Metalaxyl had similar fragmentation to those indicates in Fig. (2). For example, all R_2=CH_2OCH_3 derivatives have a base peak of m/z 45 was obtained. Despite the large number of common in the examined compound, the library search algorithm had no problem in correctly identifying the injected standard.

Figure (1): General structure of metalaxyl-fungicides

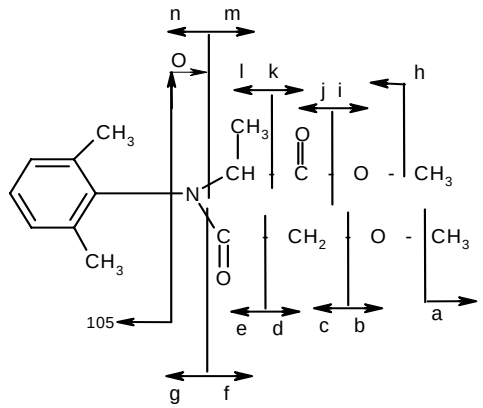


Figure (2): General fragmentation pattern for metalaxyl fungicide

Table 1: General fragmentation pattern for metalaxyl

Compound	M^+	m/z (% intensity)
Metalaxyl	279 (100%)	279(1), 249(6), 234(7), 220(11), 206 (23), 192 (13), 174 (6), 162 (9), 160 (21), 148 (10), 146 (18), 132 (18), 130 (17), 105(10), 91 (4), 77(8), 59(10), 45 (10)

In case of alkyl which group is attached to a benzene ring, preferential fragmentation occurs at a benzylic position to form a fragment ion of the formula $C_7H_7^+$ (m/e = 91). In the mass spectrum m/e = 91 loss of hydrogen from the molecular ion gives a strong peak at m/e = 91. while it may be expected that this fragment ion peak is due to the benzyl carbonium ion, evidence has been a massed which suggests that the benzyl carbonium ion actually rearranges to form the troplium ion (Pavia et al. 1977).

MS data were in good agreement with those of the hypothesis form metalaxyl., as indicated by the daughter ion spectra of protonated metalaxyl and their fragmentation scheme are presented in chart (1). (Cooke et al. 1982; Marucchini et al. 1983; Ripley 1985 and Brian1985).

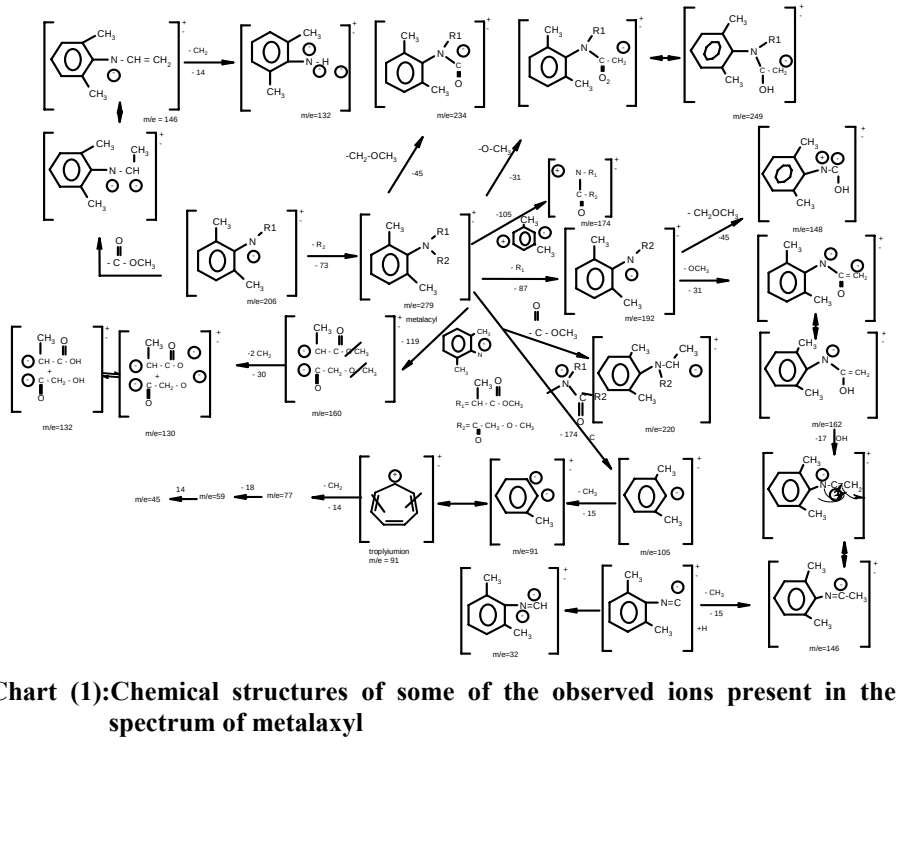


Chart (1):Chemical structures of some of the observed ions present in the spectrum of metalaxyl

Part II

Impurities of Metalaxy (2,6 dimethylaniline):

A) Estimation 2,6-dimethyl aniline in metalaxyl technical as impurities:

Retention time and data were generated on a GC as follows:

Retention time: 3.6 m.

Calculation it is found a 0.00127 g/k

FAO maximum limit 1 g/kg

The result was in the permissible limit.

B) Mass spectra

Mass spectra of 2,6 dimethylaniline which has molecular ion 121, and its relative abundance 100 (M^+)

The data fragmentation of 2,6-dimethylaniline were in agreement of (Schwinn et al., 1977; House Worth L.D. ,1987; Sanyal, and Dureja., 1992;, Sukul et al.,1992 and Dureja. et al. .,2000). Which discussed from chart (1) and Table (2) is as follows: The

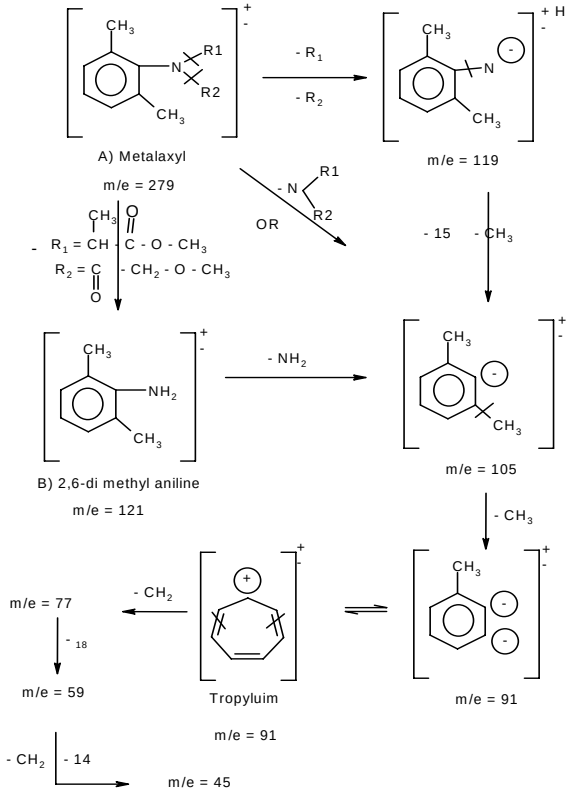


Chart 2:Chemical structures of some of the observed ions present in the spectrum of metalaxyl illustrated fragment ion of 2,6 dimethylaniline

Table 2: General fragmentation pattern for 2,6-dimethylaniline

Compound	M^+	m/z (% intensity)
2,6- di meaniline	121	105(10), 91(4), 77 (8), 59(10), 45(100)

From the previous data it can be concluded that:

The developed Method by using GC and GC/MS determination of metalaxyl and its impurity 2,6-dimethylaniline is sensitive, accurate and precise.

The present method refers to the advantage over that of previously described electron impact assay to separate the pervious compounds achieving a correct quantification of both compounds.

The procedure developed was applied to a field study on the fragmentation of metalaxyl, which shows that the main metabolites are 2,6-dimethylaniline (impurity).

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التعرف على التركيب البنائي للميتالاكسيل الخام والشوائب الرئيسية المصاحبة له

باسم السيد السيد محمد البدرى

قسم بحوث تحليل المبيدات – المعمل المركزي للمبيدات – مركز البحوث الزراعية الدقي – الجيزة

تم استخدام أحدث طريقة تحليل بجهاز التحليل الكروماتوجرافي الغازى وكذلك التحليل الكروماتوجرافي الغازى – مطياف الكتلة لدراسة الشوائب الرئيسية المصاحبة للميتالاكسيل وتم استخدام الميتالاكسيل الخام 98% لهذه الدراسة. وأظهرت الدراسة أنه نتاج . نماذج من الأجزاء المتحطمة ألمتشابهه متضمنة العديد من الروابط الأحادية.

وأظهرت أيضا أن لكل من الميتالاكسيل وشائبته زمن محدد خاصة بكل منهم من خلال عمود التحليل الشعري الغازي SE-30 وكذلك الأيونات العامة وكذلك المتعددة كشائبة الميتالاكسيل في التعرف لإثبات المركب وشائبته . وتعتبر الدراسة السابقة من أساسيات التحليل بقسم بحوث تحليل المبيدات.