Title

Studies on the Pathways of DDT by Chemical Conversion I.

Pathways of p, p'-DDT

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preliminary test is taken account of, remaining Sumithion is calculated to be less than 0.12 ppm. Thereafter Sumithion decreased rather rapidly. The time lag between the final application and the maximal content of Sumithion might indicate that Sumithion sprayed to a canopy of cocoa tree transferred gradually into the beans.

The determination of metabolic products resulting from the use of Sumithion is potentially so important from the view-point of public health. However, such methods have not yet been devised except for p-nitrocresol, one of the degradation products of Sumithion. In the treated cocoa beans p-nitrocresol was also detected, but the amount was less than 0.06 ppm.

These contents of Sumithion as well as p-nitrocresol seem to be quite negligible and completely harmless to human body from the medical viewpoint.

Acknowledgment: We wish to express our sincere thanks to Dr. L. K. Opeke, Ag. Director, and Mr. P. F. Entwistle, former Ag. Deputy Director, of West African Cocoa Research Institute for arranging the spray programs and supplying the samples of the cocoa beans. We are also grateful to Sumitomo Chemical Co., Ltd. for permission to publish this work.

Summary

The contents of Sumithion and p-nitrocresol in cocoa beans were determined after the plant had been sprayed with Sumithion.

Sumithion remaining in the beans was approximately 0.1 ppm and the content of p-nitrocresol, one of the degradation products of Sumithion, was less than 0.06 ppm.

These contents are considered to be too small to exhibit any harmful effects to human body from the view-point of public health.

Reference


Studies on the Pathways of DDT by Chemical Conversion. I. Pathways of p, p'-DDT. Yuh-Lin Chen and Hong-Ming Cheng (Pesticide Chemistry Laboratory, Department of Agricultural Chemistry, College of Agriculture, National Taiwan University, Taipei, Taiwan). Received March 1, 1965. Boryu-Kagaku, 30, 1: 1965.

11. DDT の化学変化経路に関する研究 I. p, p'-DDT の経路について 陈玉麟・郑弘命 (国立台湾大学 農学院 農業化学系 農業化学研究室) 40, 3, 1 受理

DDT は昆虫体内では酵素 DDT-dehydrochlorinase の作用で脱塩素されて DDE になることは一般に良く知られているが、最近になってから成る種の昆虫では酸化的代謝が起こり、Keltiane や DBP となることがわかった。化学的にも DDT を DDE に変える脱塩素反応は容易に行われ得るが、DDT を Kelthane へ脱酸することは簡単には行われない。著者等は DDT は TDE を原料として化学的に種々の誘導体を合成し、これら化合物間の相互関係を明らかにして、昆虫皮は動物体内での代謝研究の結果と比較し便ならしめた。本研究においては20種の p, p'-DDT の誘導体を合成し、38個の化学的変化の経路を明らかにしたが、これら化合物は iso-Acetoxy-K-3926 を除いては何れも今迄に知られているものである（Fig.1). 実験及び分析の結果は表に示した（Table 1).

著者等は更に今迄に知られている昆虫での代謝経路の外に昆虫で起こり得る新しい代謝経路 8 個を推測した（Fig. 2). この種の研究は今後の DDT 及は DDT 誘導体の昆虫皮は動物における代謝の研究に役立つもので、化学的変化の経路を昆虫における代謝のそれと比較することは極めて興味深いものと思われる。

It is well known that DDT is easily dehydrochlorinated by the action of an enzyme, DDT-dehydrochlorinase, to yield DDE in the insect body. Recently, the other metabolic products
Kelthane, an alcoholic type compound, and DBP, a ketonic type compound of DDT derivatives, were also discovered in several species of insects by Tsukamoto and Perry and Miller respectively. It is concluded that, in general, the metabolic pathways of DDT are now considered to be two; one to DDE by the dehydrochlorination and the other to Kelthane or DBP by the oxidative metabolisms in insects.

The dehydrochlorination of DDT to DDE by chemical method is also very easy. In fact, DDE is obtainable by simply treating DDT with alcoholic alkaline solution without any difficulty. On the other hand, it is not so simple to prepare Kelthane from DDT by the chemical conversion. No method has been demonstrated to convert DDT to Kelthane in a single step. It is necessary to dehydrochlorinate DDT to DDE, then chlorinate DDE to Cl-DDT, acetoxylate Cl-DDT to Acetoxy-DDT, and finally hydrolyse Acetoxy-DDT to Kelthane.

The purpose of this investigation is to synthesize some different derivatives of DDT by chemical conversion from DDT or TDE as the starting material and to clear up the chemical relationship among these compounds. It is of interest to compare the relationship between the chemical conversion and metabolic pathways known in insects and animals.

Twenty compounds were prepared and thirty-eight pathways of chemical conversion were identified in this investigation. These pathways are shown in Fig. 1, together with that of the study of metabolic fate of DDT in insects as shown in Fig. 2. The results of experiments and analytical data are tabulated in Table 1.

The chemical names of compounds and the abbreviation used in this paper are given below:

1. CI-DDT : 1, 1-bis (p-chlorophenyl)-2, 2, 2-tetrachloroethane.
3. DBP : 4, 4'-dichlorobenzenophenone.
4. DDA : bis (p-chlorophenyl)-acetic acid.
5. DDE : 1, 1-bis (p-chlorophenyl)-2, 2-dichloroethylene.
6. DDT : 1, 1-bis (p-chlorophenyl)-2, 2, 2-trichloroethane.
8. iso-DDT : 1, 1-bis (p-chlorophenyl)-1, 2, 2-trichloroethane.
9. TDEE : 1, 1-bis (p-chlorophenyl)-2-chloroethylene.
10. TDE : 1, 1-bis (p-chlorophenyl) -2, 2-dichloroethane.
11. FW-152 : 1, 1-bis (p-chlorophenyl)-2, 2-dichloroethanol.
13. DME : 1, 1-bis (p-chlorophenyl)-ethane.
14. DDM : 1, 1-bis (p-chlorophenyl)-2-chloroethane.
16. iso-DMC : 2, 2-bis (p-chlorophenyl)-ethanol.
18. DMC : 1, 1-bis (p-chlorophenyl)-ethanol.
21. DCPM : bis (p-chlorophenyl)-methane.
22. DBH : bis (p-chlorophenyl)-methanol.
23. DCMC : 1, 1-bis (p-chlorophenyl)-2-chloroethanol.
24. iso-Kelthane : 2, 2-bis (p-chlorophenyl)-1, 1, 2-trichloroethanol.
25. iso-TDE : 1, 1-bis (p-chlorophenyl)-1, 2-dichloroethane.
26. iso-FW-152 : 2, 2-bis (p-chlorophenyl)-1, 2-dichloroethanol.
27. iso-DDM : 1, 1-bis (p-chlorophenyl)-1-chloroethane.
28. iso-DCMC : 2, 2-bis (p-chlorophenyl)-2-chloroethanol.

The authors wish to express their sincere thanks to Prof. Dr. R. Yamamoto, Tokyo University of Agriculture, Prof. Dr. Y. Oshima, Kyushu University, and Dr. M. Tsukamoto, Osaka University, Japan, for their encouragements and valuable suggetions. This investigation was carried out in cooperation with Prof. Dr. R. Yamamoto, Tokyo University of Agriculture, and Dr. M. Tsukamoto, Osaka University, Japan.
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out with the support of a grant-in-aid from the National Council on Science Development. DDT and TDE (Rhothane) used in this experiment were furnished by the Agricultural Chemical Works, Kaohsiung, and Elephant Trading Co., Ltd., Taipei, Taiwan, respectively, to which thanks are due.

**Experimental**

DDT used in this experiment was recrystallized.
twice with ethanol from the technical grade produced by the Agricultural Chemical Works, Kaohsiung, Taiwan. The recrystallized product showed mp. 107−108°. TDE was extracted from the commercial product of Rhothane WP-50, the product of Rohm and Haas Co., U.S.A., and recrystallized from ethanol, showing mp. 110−111°. Both compounds were proved to be paper chromatographically pure.

Pathways 1 and 17: Prepared from DDE or TDEE according to the method of Bergmann and Kaluszyner.6)

Pathways 2 and 16: Kelthane or FW−152 was refluxed for 3 hrs. with an excess of thionylchloride in a dry benzene solution.

Pathways 3 and 21: Cl-DDT or iso-DDT was refluxed with mercuric acetate in an acetic acid solution for 90 minutes. The mp. of Acetoxy-TDE was reported to be 129−131°.12)

Pathway 4: Crude Kelthane was refluxed with acetic anhydride and few drops of conc. sulfuric acid for 3 hrs.

Pathways 5 and 8: Oxidized from DDA or DDE with potassium dichromate and conc. sulfuric acid in an acetic acid solution. The mp. of DBP was reported to be 146−147°.8)

Pathways 6 and 7: The product was obtained by treating Acetoxy-DDT or Kelthane with alcoholic potassium hydroxide solution.

Pathways 9 and 10: DDA was prepared from DDT or DDE by the method of Grumitt et al.6)

Pathways 11 and 18: Cl-DDT or iso-DDT was reduced with metallic sodium in a boiling solution of absolute ethanol. The mp. of DBH was reported to be 92.8−93.3°.13)

Pathways 12, 14, 19, and 25: Dehydrochlorination was carried out with acetic anhydride, a small quantity of pyridine and sodium acetate.

Pathways 22 and 34: FW−152 or iso-DMC was refluxed for 3 hrs. with acetic anhydride, a small quantity of pyridine and sodium acetate.

Pathways 23 and 38: iso-Acetoxy-K−3926 or DBP was treated with metallic sodium in a boiling solution of absolute ethanol. The mp. of DBH was reported to be 92.8−93.3°.13)

Pathway 26: iso-DMC was treated with thionylchloride and pyridine at room temperature. The mp. of DDM was reported to be 51−53°.14)

Pathways 27 and 33: Synthesized from DDA, methanol or ethanol and a small quantity of conc. sulfuric acid. Reaction was carried out at reflux for several hours.11) The mp. of ethyl ester was reported to be 87.5−88°.15)

Pathways 28 and 29: Attempts to reduce DDA or its ester with metallic sodium or with zinc and acid were not successful. These were finally obtained by the reduction with lithium aluminum hydride16). The mp. of iso-DMC was reported to be 98.5−99.5°17).

Pathway 30: iso-Acetoxy-K−3926 was refluxed for 2 hrs. alcoholic potassium hydroxide solution.

Pathway 31: DDT was reduced with zinc granule and conc. hydrochloric acid in a solution of ethanol or acetic acid. The mp. of K−3926 was reported to be 54−55°.10) Besides K−3926, some by-products with higher melting points were obtained. But these were not further investigated.

Pathway 32: Prepared by the Grignard reaction from methyl iodide, metallic magnesium, and DBP according to the method of Grumitt et al.10)

Pathway 35: DDA was treated with barium hydroxide in an ethylene glycol at above 190° according to the method of Cristol and Haller.18)

Pathway 36: DBP was reduced with zinc granule and conc. hydrochloric acid by refluxing for about 10 hrs. in a solution of acetic acid. It was also prepared by the Clemmensen reduction17). Besides DCPM, a higher melting product (mp. 213−215°) was obtained in 13−20% yield from both methods. The compound was identified to be 4, 4', 4", 4"'-tetrachlorobenzopinacol(Analysis, Calcd. for C₈₄H₂₄O₂Cl₄: Cl, 28.13%found : Cl, 27.88%, 28.37%).

Pathway 37: DBH was reduced with zinc granule
and conc. hydrochloric acid by refluxing for 8 hrs. in an acetic acid solution.

Discussion

Besides several metabolic pathways identified by Tsukamoto, (Fig. 2, pathways a~g) and by Perry and Miller, (Fig. 2, pathways h~i), possible metabolic pathways in insects were added by the authors (Fig. 2, pathways j~q). Although the pathway from iso-DDT to iso-Kelthane (Fig. 2, pathway K) was already proposed by Tsukamoto, it seems to be possible to metabolize to DDE (Fig. 2, pathway j) simultaneously.

Owing to the lack of facilities, several important reactions such as the catalytic hydrogenation under the high pressure still could not be performed in this investigation. Moreover, many possible chemical pathways were not successful in this experiment, such as the chlorination of DME to iso-TDE, DMC to iso-DDM, acetoxylation of DMC to Acetoxy-K-3926, and reduction of TDE or DDM to K-3926 etc. Further studies are still needed to complete this investigation.

Many compounds were prepared in this investigation. But only one compound, iso-Acetoxy-K-3926, seems to be a new compound.

DDA, Acetoxy-DDT, Kelthane, and DDE were easily converted to DBP by chemical methods. But DDT itself could not be oxidized to DBP with potassium dichromate. The result is well agreed with that of the metabolic pathway in insect.

It is very interesting to compare the chemical pathways with that of the metabolic pathways in insects, and the results obtained are seemed to be very helpful in the future study of metabolic fate of DDT or related compounds in insects and animals.

Summary

It is well known that DDT is easily dehydrochlorinated by the action of an enzyme, DDT-dehydrochlorinase, to yield DDE in the insect body. Recently the other oxidative metabolic products Kelthane and DBP were also discovered in several species of insects. The dehydrochlorination of DDT to DDE by chemical method is also very easy. On the other hand, it is not so simple to prepare Kelthane from DDT by the chemical conversion. The purpose of this investigation is to synthesize some different derivatives of DDT by chemical conversion from DDT or TDE as the starting material, and to compare the chemical relationship among results of metabolic studies in insects and animals.

Twenty compounds were prepared and thirty-eight pathways of chemical conversion of p,p'-DDT were identified in this investigation (Fig. 1). But only one derivative, iso-Acetoxy-K-3926, seems to be a new compound. The results of experiment and analytical data are tabulated in Table 1. Besides nine metabolic pathways identified by the former investigators, eight possible metabolic pathways which might be occurred in insects were also proposed by the authors (Fig. 2).

It is very interesting to compare the chemical pathways with that of the metabolic pathways in insects, and the results obtained are seemed to be very helpful in the future study of metabolic fate of DDT or related compounds in insects and animals.

References

12) Craig, W. E. Shropshire, E. Y. and Wilson, H.
Fig. 1 Scheme of pathways of $p,p'$-DDT by chemical conversion
Fig. 2 Scheme of metabolic pathways of \( p,p'\)-DDT and its derivatives in insects

1. *Drosophila melanogaster*
2. *Drosophila virilis*
3. *Musca domestica*
4. *Bletella germanica*
5. *Pediculus humanus humanus*
Comparison of Synergistic Action of Anti-resistant DDT and DMC with DDT Against the Common House Fly Evaluated by the Impregnated Filter Paper Method. Studies on the Biological Assay of Insecticides. LIII. Sumio NAGASAWA and Michiyo Sstarik (Ihara Agricultural Chemicals Institute, Shimizu, Shizuoka). Received March 29, 1965. (with English Summary 61)

12. イエバエをもいる液紙法によって観察した DDT に対する anti-resistant DDT と DMC の協力効果の比較 研究を含む生物試験に関する研究。第 53 西 日本農業研究所 40, 3, 29 受理

DDT に対する anti-resistant DDT の協力効果と DMC のそれらを、イエバエをもいる液紙法によって比較した結果、致死を決定する要因として塩素気を用いた場合、anti-resistant DDT の場合ほとんど同様の強度性を有するが、DMC の場合は塩素気より致死時間の方がより重要で、また塩素気だけを考慮した場合は、両物質においてほとんどとも」といったことが明らかにされた。したがって、液紙法の相対協力効果をプロット平面の直線距離で表すと、致死時間が長い間は anti-resistant DDT の DDT に対する協力効果は、DMC のそれより大きいか、致死時間が短くにつれてその相対値は小さくなり、理論上致死時間が 312 分になったとき、両物質の強力性を示すと、いうことが可能である。

実験材料および方法

実験材料: この実験に用いた p,p'-DDT は、メタノールで再結晶した m.p. 108°C の試料である。以下の DDT として、その強度を有する anti-resistant DDT (N,N-di-n-butyl-p-chlorobenzene sulfonamide) よりも DMC (1,1-bis-p-chlorophenyl methyl carbinol) は、日本農業株式会社生物研究所から提供をうけた research grade の試料である。イエバエは豆腐化能力によっててその幼虫期を、砂糖と水によって成虫期を経て、いわゆる高発性体称される耐毒労働系統で、実験には羽化後 4〜5 日目の個体群を用いた。本実験の挿入方法による DDT の LD₅₀ は約 7μg/μg、測定の標準性の感度性にくらべると、その値はかなり高いようである。

実験方法: DDT と anti-resistant DDT あるいは DMC を等量混合し、アセトンでこの 12.5, 25, 50 および 100μg/m² の希釈溶液をつくり、その 1 cc を直径 9 cm の液紙（東洋液紙 No. 2）に一様に滴下し、乾燥のことを兼ねて直進のベトリ皿をかぶせ、これにイエバエ約 30 匹を導入、所定の 80,100 および 160 分間観察した。観察後は直径 9 cm、高さ 5 cm のガラス容器に封じ、縁をかぶせた後その上から脱脂綿にひたした稀釈牛乳を戻としてあてた。観察を終えた時からかえて、24時間後にその死亡数をかぞえた。別に対照区としてアセトンのみを 1 cc 調製して、同様の実験をおこなった。処理区対照区とも実験は 2〜3 に再繰り返し、その結果を集計した。観察、実験ともに 25°C、関係温度約60%の環境条件下で施行した。