

EXHIBIT 39

FILED UNDER SEAL

1130/2

Dr. F. E. Kohn,
Assistant Laboratory Director,
Industrial Bio-Test
Laboratories Inc.,
1810 Frontage Road,
Northbrook, Illinois,
U. S. A.

JSQ/EM

*California Chemical Co.
Ortho Division
Lucas Street Northbrook
Richmond California
U.S.A.*

17th February, 1964.

Dear Dr. Kohn,

Dr. Swan has asked me to comment on the difficulties which you have encountered in the determination of paraquat in rat and dog urine.

I do not know whether you have been using the same method as we do here, so I enclose a copy of our procedure. We have recently subjected this method to a searching test as we have been using 14C labelled paraquat to investigate the extent of metabolism in the rat after oral dosing. We have certainly found several sources of error. Paraquat disappears fairly rapidly from urine, presumably due to microbiological attack, unless the urine is frozen and kept in the dark as soon as it is passed. We have a rat metabolism case which achieves this, but in proposed work in dogs we think it will be necessary to take urine samples by catheter. Another source of error appears to be the adsorption of paraquat on precipitates, probably mainly calcium phosphate, which form in urine. There also seems to be a slow loss of paraquat if the trichloroacetic acid solution after protein precipitation is kept. When care is taken to avoid these sources of error, we find an exact equivalence between the paraquat and radioactivity measured in urine, indicating that no metabolism has taken place in the rat.

We have no doubt that paraquat is associated with protein in urine, and treatment of the urine with trichloroacetic acid is necessary in order to obtain satisfactory recoveries. I find it difficult, however, to understand how this can be overcome by making the urine just alkaline, if immediately afterwards it is made strongly acid with trichloroacetic acid.

I am very puzzled by your observation that urine, which apparently contains no paraquat when analysed, will give a blue colour when made alkaline. I have consulted Plant Protection Ltd. and I am informed that if 0.1 M paraquat is mixed with 2N NaOH a blue colour will slowly develop, more readily seen if air is excluded, but they would not expect this to occur at the concentrations encountered in urine. We have not managed to obtain a blue colour with such concentrations.

2.

We have been disturbed by your comments on the determination of paraquat in urine and we should be most interested to know whether you have any further information.

Yours sincerely,

(J. C. Gage)

Copy to: Dr. J. C. Calandra