



# THE RHEUMATOID DISEASE FOUNDATION

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*President*

## IMIDAZOLE COMPOUNDS FOR THE TREATMENT OF RHEUMATOID ARTHRITIS

Presentation by:

John R.A.Simoons, Ph.D. at 1984 fall convention AAMP, Dallas Tx.

Thank you, Dr. Prosch for the introduction.

Ladies and Gentlemen:

It is indeed a pleasure and an honor for the invitation to present a talk to such a select audience of physicians and scientists attending the fall convention of 1984 of the American Academy of Medical Preventics in Dallas, Texas.

My talk will discuss the use of Imidazole compounds for the treatment of rheumatoid arthritis, based on their anti-protozoal activity.

Parasitic infections, which include the free living-amoeba are not limited to tropical and subtropical countries as previously thought but encountered throughout the world. Of the more than one hundred drugs which have been or are still being used for treating such diseases as Amebiasis, Ancylostoma, Ascaris lumbricoides, Filariasis, Giardiasis, Leishmaniasis, Malaria, Mites, Schistosomiasis, Trichomoniasis, Trypanosomiasis etc. I would like to discuss only the IMIDAZOLE compounds because these are by far the most effective and probably the safest, when used with caution.

My close relationship with Dr. Wyburn-Mason which started in October 1975 until his death in June 1983, during which time we met often twice a year to discuss his revolutionary new concept of an amebic infection as the etiology of rheumatoid diseases and other collagen and auto-immune diseases, convinced me that he was right. We reviewed hundreds of case histories of patients from all over the world who visited him for treatment, after years of unsuccessful conventional therapy. Of the various anti-amoebic compounds which he used for treatment and which are listed in

- 2 -

THE RHEUMATOID DISEASE FOUNDATION IS A PROJECT OF  
THE ROGER WYBURN-MASON & JACK M. BLOUNT FOUNDATION  
FOR THE ERADICATION OF RHEUMATOID DISEASE

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his books and various publications on his research of more than thirty years, we studied the IMIDAZOLE compounds more extensively.

In spite of earlier reports that rheumatoid arthritis responded favorably to the treatment with anti-amoebic compounds, no one saw the link of these infections and rheumatoid disease.

In 1951, Dr. Emmanuel Rappaport treated four patients with intestinal amebiasis who also had rheumatoid arthritis with DIODOQUIN and obtained excellent results. He isolated *Entamoeba histolytica* from the stool and suggested that arthritis in these patients was probably due to sensitization to a toxin elaborated by the amoeba (Annals Internal Medicine, 1951, Vol. 34, 1225-1231)

Dr. Rappaport came very close but failed to see the connection. Thirty years later, Dr. Robert Bingham applied Diodoquin in 204 patients for the treatment of rheumatoid arthritis and reported a 93% remission.

In 1970, Abdul Rabbo applied a new anti-amoebic Imidazole compound, listed as BT 985 of MERCK A.G. in a few patients with acute lupus erythematosus and obtained complete remission which lasted six months. Encouraged by these results he then tried the drug in 12 patients with active rheumatoid arthritis who had not responded to other remedies and were severely ill. He reported a 90% success rate and described the results as "dramatic" in the American J. of Trop. Med. Hyg. 1972, 75, 64-66. Abdul Rabbo also failed to see the link of an amoebic infection and the disease.

Research chemists and pharmacologists have long established that there is a certain relationship between the chemical structure and biological activity of a compound and that this can be modified by introducing certain radicals or adding or changing substitutions which could influence their mechanism of action. Biological half lives, toxicity, site of absorption, metabolic fate and elimination can all be affected by chemical manipulations. As an example we may refer to the corticosteroids of which the basic structure consists of three 6-carbon rings and a single 5-carbon ring, the Cyclo-pentano-phenantrene nucleus. Our own system synthesises corticosteroid hormones from dietary cholesterol.

IMIDAZOLE COMPOUNDS IN R.A.

John R.A.Simoons, Ph.D.

Substitution of a hydroxyl group at C-11 in the basic compound CORTISONE, produces Hydrocortisone(Cortisol)the major glucocorticoid secreted by the adrenals in man.A ketogroup at C-3 of the steroid molecule is essential for glucocorticoid activity. An oxy or hydroxyl group at C-11 is indispensable for anti-inflammatory activity.We could prepare short acting(hydrocortisone) intermediate acting(prednisone and prednisolon) and long acting(dexamethasone) corticosteroids by certain substitutions at certain positions.Another example are the antihistamines in which we could modify their sedative or anti-parkinson activity in Orphenadrine by changing the position of the methylgroup.

The IMIDAZOLE compounds remained of great interest to research chemists because they exhibit specific anti-microbial activity against a wide range of micro-organisms unaffected by the more widely used antibiotics.We conducted an extensive literature search of a large number of synthesized IMIDAZOLE derivatives and tried to find the relationship between their chemical structure and biological activity against certain micro-organisms. Their activity is listed as antifungals,antiprotozoals,anti amoebics,antiparasitics etc.Based on laboratory and clinical findings it appeared that compounds with a substitution at the ONE position were more anti-protozoal in action.A short aliphatic substitution produced a weaker anti-protozoal activity than more complex cyclic groups.The NITRO group at the 4 or 5 position also seemed to increase the toxicity.

Dr.Wyburn-Mason was, of course, more interested in Imidazole compounds which were available for systemic use and which he could prescribe for his patients.

METRONIDAZOLE(Flagyl) S l i d e # 1

This compound satisfied the substitution at the ONE position of a 2-hydroxy-ethyl group, which is a short aliphatic chain and also has a nitro group at the FIVE position.

First cases treated by Wyburn-Mason were negative in the dose which he applied until Dr.Blount informed him of his dramatic success at a much higher dose with, however, more side-effects. By modifying the dosage to a treatment of two consecutive days per week of about 2 g per day for 6 - 12 weeks, they obtained

IMIDAZOLE COMPOUNDS IN R.A.

John R.A.Simoons, Ph.D.

very favorable results and less side-effects.

We know that these compounds produce an induction of liver enzymes which could be prevented by a 2day per week dosification. The Herxheimer reaction develops also less severe and patients could tolerate the treatment much better.

TINIDAZOLE (Fasigyn) S l i d e # 2

This compound is widely used in several European countries to treat trichomonal infections in women. It has a 2-ethylsulfonyl-ethyl substitution at the ONE position and a NITRO group at the FIVE position. Just like Metronidazole it also has a METHYL group at the TWO position.

Dr. Wyburn-Mason called this his most favorable compound because he saw less side effects, although several patients failed to improve. He believed that these cases could then be treated with other Imidazole compounds to which the causative organisms were more sensitive.

LEVAMISOLE , S l i d e # 3

This compound is listed as an anthelmintic and has been most widely tested by Janssen Pharmaceuticals and Ortho since 1975. It has a beta thiazole group at the ONE and TWO positions and a tetra-hydro-6-phenyl group at the FOUR position. The compound has a narrow therapeutic range just like Penicillamine and Methotrexate with which it is being compared.

Efficacy and side effects are shown in S l i d e # 4

The side-effects increase as the dose is increased as is shown in the next slide, S l i d e # 5. The lower dose also showed a lower effectiveness. Clinical studies of this compound were discontinued after the FDA rejected further evaluation in view of serious side-effects.

TIFLAMIZOLE, S l i d e 6

Synthesized by Dupont, this compound has aliphatic Fluor and Cyclic Fluor substitutions at the TWO, FOUR and FIVE positions and has no anti-protozoal activity. In a series of phase one studies in several medical centres the compound showed a high degree of severe side effects and no efficacy in the treatment of rheumatoid arthritis. We believe that further evaluation will be halted.

CLOTRIMAZOLE, S l i d e 7

The compound is a tritylimidazole with a bis-phenyl-2chloro-phenyl substitution at the ONE position. There is no NITRO group in the formula and we consider this one of the most potent anti-protozoal compounds which has been extensively tested since 1972. (Postgraduate Medical J., July 1974, Vol. 50 1-108 and Curr. Med. Res. Opin. 1977, 5, 169-178.

Clotrimazole is listed as a broad spectrum antimycotic agent. It is fungistatic and fungicidal against pathogenic fungi, moulds, protozoan and certain bacteria and therefore not only covers the spectrum of activity of the polyenes (Amphotericin B, Nystatin and Candicidin) but also that of Griseofulvin.

Clotrimazole is well absorbed from the gastro-intestinal tract as high as 90% within one hour and peak blood levels are reached around three hours, maintaining serum levels of over 1 mcg/ml for more than 24 hours after a single oral dose of 40mg/kg.

Pharmakokinetic studies with <sup>14</sup>C labelled Clotrimazole were conducted in ten healthy volunteers. The compound is almost completely metabolized and the metabolites are excreted in the bile. Only 10% of the metabolites are excreted in the urine.

In spite of reports on an increased Cortisol blood levels after oral administration of 80mg/kg/day, Bayer reported that there was no evidence of interference with adrenal function from the measurements of ketosteroid excretion in 13 healthy volunteers after 28 days of oral administration at a dose of 40mg/kg/day. The LD/50 in mice is about 800 mg and 1500 mg in rabbits.

Side-effects at doses of 40mg/kg/day during 28 days include Gastro-intestinal disturbance, Nausea, Vomiting, Diarrhoea and Anorexia in about 8% of the patients. Also reported are lethargy, depression and irritability in a few patients.

WYBURN-MASON was the first to use CLOTRIMAZOLE in the treatment of rheumatoid arthritis as reported in The Lancet of Febr. 28, 1976, 489. He used an initial dose of 100mg/kg/day but after severe gastro-intestinal disturbance in several patients, he continued at a lower dose of 25mg/kg/day for up to 12 weeks. He described symptoms of a severe Herxheimer reaction which the patients developed and believed that this is typical of reactions seen in patients with parasitic diseases treated by drugs which kill the causative organisms.

IMIDAZOLE COMPOUNDS IN R.A.

John R.A.Simoons, Ph.D.

Active disease disappeared in 3 - 28 days. After 6 weeks ESR and blood count became normal. In 4 - 6 months the albumin/globulin ratio and the electrophoretic pattern returned to normal. During the follow-up period of up to 15 months there was no return of disease activity. (The Lancet, Febr. 28, 1976, 489: Wyburn-Mason)

BAYER U.K., obtained approval from the British Committee on the safety of drugs the equivalent of our FDA to evaluate Clotrimazole in a controlled double blind study after Wyburn-Mason's report.

WOJTULEWSKI, conducted a controlled double-blind study in 47 patients with active rheumatoid arthritis during eight weeks in which Clotrimazole was compared with a standard non-steroidal anti-inflammatory agent, Ketoprofen (Orudis, Motrin). The dose for the first week was 40mg/kg/day followed by 80mg/kg/day for seven weeks. Slide 8 shows the efficacy of Clotrimazole treatment versus Ketoprofen.

The incidence of side effects as expected from this high dose is shown in slide 9.

The effect on the white blood cell count is shown in slide 10.

We visited with Dr. Wojtulewski after the clinical study and he informed Dr. Wyburn-Mason and myself that he obtained better results and less side-effects at a dose of 1g/day for up to 32 weeks in an open study. We received several data from him and the next slide, slide 11, indicates the results of his open study in a continuation of the first study.

The Rheumatoid Disease Foundation has sponsored the first double-blind clinical study of Clotrimazole against Placebo which is now being conducted by the Bowman Gray School of Medicine in Winston-Salem, N.C. Miles Pharmaceuticals, a subsidiary of BAYER provided the Clotrimazole 250 mg tablets and identical Placebos and Dr. Robert Turner, Professor of Medicine and Chief section on rheumatology is the chief investigator. The IND has been approved by the Food and Drug Administration as well as the protocol for the study. Slide 12.

I would like to point out that the Food and Drug Administration requires that a drug is effective and safe for its intended use in humans before an NDA will be approved. The safety of Clotrimazole for use in humans has been approved by the FDA

IMIDAZOLE COMPOUNDS IN R.A.

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because they approved NDA# 18-713 for Mycelex in 1984 which is an oral Clotrimazole preparation, submitted by Miles Pharmaceuticals. We hope to establish "effectiveness" of Clotrimazole in rheumatoid arthritis in the coming months by our clinical evaluation at Bowman Gray and confirm Wyburn-Mason's findings.

The Rheumatoid Disease Foundation would, of course, also try to confirm Wyburn-Mason's hypothesis of a protozoal causation in rheumatoid disease and establish the mechanism of action of anti-amoebic compounds.

In 1978 we conducted "in vitro" studies comparing the anti-protozoal activity of several Imidazole compounds against cultures of *Naegleria fowleri*. We found Clotrimazole to be the most potent of the Imidazole compounds tested. Metronidazole was the weakest. Tinidazole was not much better. Several published reports confirm that Clotrimazole is amoebicidal against strains of *Naegleria* and *Acanthamoebae* and Jamieson established that it was amoebicidal to *Naegleria fowleri* at concentrations of 0.1 to 1.0 mcg/ml which is about the serum levels obtained after oral administration. Our Tony Chapdelaine is conducting a series of "in vitro" experiments at Vanderbilt University with Dr. Neff to determine the anti-amoebic activity of several compounds. His initial reports established that Clotrimazole is amoebicidal to *Acanthamoeba Culbertsoni*.

Reports by Cline published in *J. Protozoology*, 1983, Vol 30 pages 387-391 compared 16 different antibiotics and Imidazole compounds in various concentrations against *Naegleria fowleri* and *Naegleria gruberi* and concluded that Clotrimazole and Amphoterecin B were the most active drugs in inhibiting growth of these *Naegleria* species.

It has been reported in studies by Bayer in their investigator's manual on Clotrimazole that this drug acts by damaging the permeability barrier of sensitive organisms, possibly through reaction with the cell membrane's phospholipid layer.

In several studies with LEVAMISOLE, evidence of stimulation of cell-mediated immunity showed a correlation between such changes and pain relief. Scientists believe that rapid deployment of PMNL (Poly-morpho-nuclear-leucocytes) function is very important in resistance to infection.

A drug should therefore not impede vital functions of the reticuloendothelial system, i.e. reducing the migration to the site of infection, phagocytosis or killing of the invading organisms by the PMNL. A recent "in vitro" study by Rowan-Kelly tested five Imidazole compounds to determine their effect on the modification of PMNL function (Int. J. Immunopharm. 1984, vol. 6, 4, 289-300).

Slide # 13, showed that the highly substituted Imidazole compounds, Clotrimazole, Miconazole and Ketoconazole caused a significant dose-dependent inhibition of the PMNL chemotaxis and that Clotrimazole reduced this function to 11.2% of the control at a concentration of 20mcg/ml. Metronidazole and Tinidazole did not effect the rate of migration. I would like to point out that serum or tissue levels of Clotrimazole will never reach a level of more than 2 mcg/ml during treatment at which level it will also not effect the migration. The study also indicated, however, that Clotrimazole did not effect the bactericidal nor fungicidal activity of PMNL function, contrary to Ketoconazole and Miconazole.

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CITIZENSHIP: The Netherlands

EDUCATION: Prince Hendrik Lyceum B.S.Chemistry 1932-1937

Doctorandus (M.S.) Magna cum laude, Biochemistry  
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May 1962; Thesis "Formulation and Experimental evaluation  
of sustained release medication based on the  
principle of delayed diffusion" (Scientific Communi-  
cations Vol. XII, Jan. 1963, 206, 1-111, Haarlem, Holland)

PROFESSIONAL EXPERIENCE:

- 1) Director Department of Pharmacology at Brocades  
Research Institute, Haarlem, the Netherlands from  
1958 - 1962.
- 2) Technical director and International Supervisor  
Organon International 1963 - 1968
- 3) Director Research & Development, Organon USA,  
West-Orange, N.J. 1968 - 1980

U.S. Patents: # 3,192,774 July 6, 1965: Apparatus for determining  
the release rate of drugs with delayed action and  
"in vivo" correlation.

# 3,322,633 May 30, 1967: Pharmaceutical formulations  
with sustained release.

# 4,154,820 May 15, 1979: Compositions containing  
Alkali Metal Sulfate Salts of Conjugated Estrogens.

LANGUAGES : Dutch, English, German, Spanish, Portuguese

October 10, 1984



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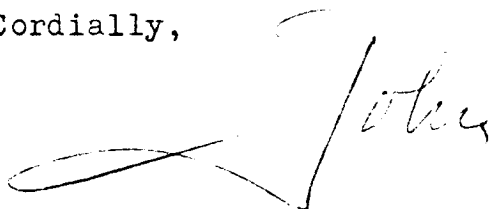
October 10, 1984

Mr. Perry A. Chapdelaine, Sr.  
Rt. 4, Box 137  
Franklin, TN 37064

Dear Perry:

I hope that the enclosed Curriculum Vitae and Synopsis and photo are what you requested. I had very little time because I must still pack for our trip tomorrow morning to N.J. but hope that you can use the enclosed information. I hope to receive details of the meeting as soon as I arrive in N.J. and I may call you if required. I will project 16 slides or may reduce it if time runs out and my talk will be for 10 minutes and 5 minutes to answer questions. I will arrange for the airline ticket but could you share a room with me or Jack with me and please make the reservations. I hope to promote our Foundation to the best of my ability but you are the most qualified PR man and I am happy that you will be there. It is of course an honor that Jack requested me to share the panel with him. Thank him for me and looking forward to hearing from you soon.

Cordially,



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