THE KAISER WILHELM INSTITUTE FUR MEDIZINISCHE FORSCHUNG HEIDELBERG

COMBINED INTELLIGENCE OBJECTIVES SUB-COMMITTEE

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NOTES ON THE KAISER WILHELM INSTITUT FÜR MEDIZINISCHE FORSCHUNG AT HEIDELBERG

Reported by

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from notes by

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CIOS BLACK LIST ITEMS

24 - Medical

COMBINED INTELLIGENCE OBJECTIVES SUB-COMMITTEE
G-2 Division, SHAEF (Rear) APO 413

TABLE OF CONTENTS

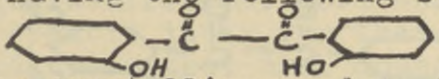
NOTES ON THE KAISER WILHELM INSTITUTE FÜR MEDIZINISCHE FORSCHUNG AT HEIDELBERG.

1. Introduction: The following report is based on information obtained by Major Cortez F. Enloe, Chief, Medical Branch, Morale Division, U. S. Strategic Bombing Survey, in the course of an investigation of Target No. 24/17a, supplemented by information derived from an interrogation of Dr. Richard Kuhn by Lt. Saffer, U.S.N.R., representing the Navy Technical Mission, ETO.

2. This important target was found intact, neither its facilities nor operation having been impaired by military activity in this area. The majority of the scientific staff of the Institute had remained and were available for interrogation. Sections of the Institute had been established at Tauberbischofsheim, and at Mudau in the Odenwald.

3. The most important and fruitful source of information encountered at this target was the director of the Institute, Professor Richard Kuhn, former Nobel Prize winner and outstanding biochemist. In the course of interrogations lasting several days, he provided information on the nature, properties and therapeutic value of various substances which he had been investigating during the war.

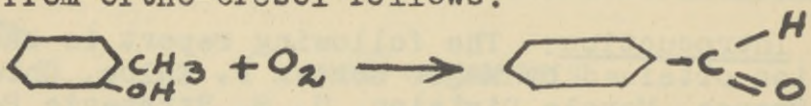
4. 2,2' dioxybenzyl of Salicil: This is a substance developed by Professor Kuhn and Dr. Leonhard Birkofer for the treatment of all types of coccal infection and diseases of the kind in which at present penicillin is indicated. This compound is prepared from Salicil, a derivative of salicylic acid, having the following structural formula:



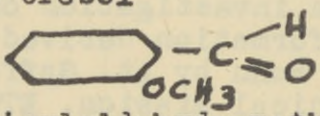
Salicil is a fine crystalline powder, yellow in color, with a melting point of 213° C. It is insoluble in water and acid solutions, but is soluble up to ten percent in slightly alkaline bicarbonate solutions and in 10% boric acid.

5. Salicil has no bactericidal action, but it has been possible to prepare two derivatives of this compound, both of which have been found to possess considerable bactericidal activity. The first of these is di-brom-salicil (Code No. 3065), prepared by brominating the original compound; the second is tetra-chlor-salicil (Code No. 3014), obtained by treating salicil with chlorine. The method of

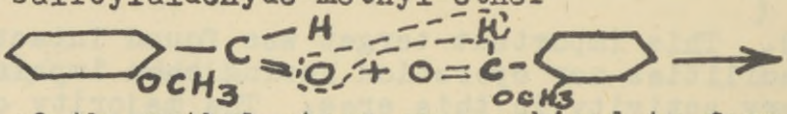
preparation of these compounds is described in detail in the attached translation of a paper published by Kuhn, Birkofer, and Möller. A graphic presentation of the synthesis from ortho-cresol follows:



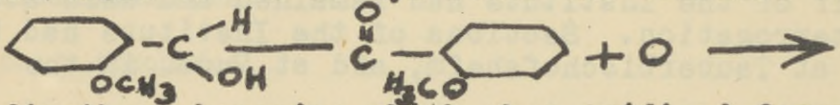
ortho-cresol salicylaldehyde



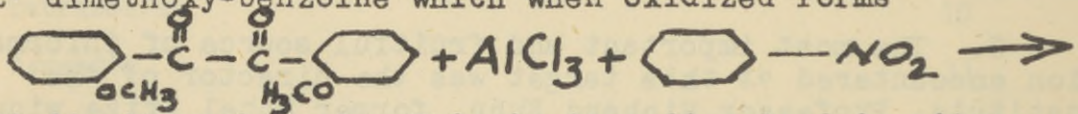
methylated to salicylaldehyde-methyl-ether



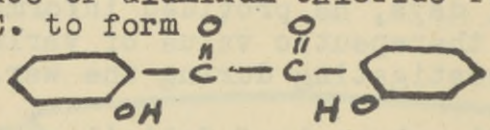
two molecules of the methyl-etner are combined to form



2,2' dimethoxy-benzoine which when oxidized forms

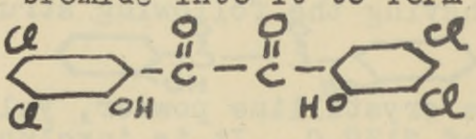


2,2' dimethoxy-benzyl. This is then dissolved in nitro-benzol in the presence of aluminum chloride for 6 to 7 hours at 50 to 60° C. to form

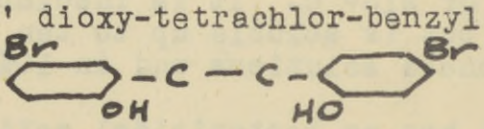


2,2' dioxy-benzyl

This substance is then dissolved in glacial acetic acid and halogenated by passing chlorine through the solution or by titrating the bromide into it to form the desired product



2,2' dioxy-tetrachlor-benzyl, or 3014



2,2' dioxy-dibrom-benzyl, or 3065

6. Salicil impedes the growth of yeasts and in contrast to salicylic acid is effective regardless of the pH of the solution. In a dilution of 1:100,000, the growth of yeasts is completely suppressed. When applied in the same manner to staphylococci, salicil is much less effective;

and the longer the duration of the experiment, the less effective is the suppressive action. Introduction of the two bromine atoms into the compound decreases the effectiveness of salicil against the yeast but, according to Kuhn, increases significantly its bacteriostatic activity. Di-brom-salicil in a dilution of 1:500,000 and even of 1:1,000,000 exceeded in effectiveness the action of sulfonamides. Sulfathiazole, for example, maintains its bactericidal activity against the strains employed up to a dilution of 1:30,000.

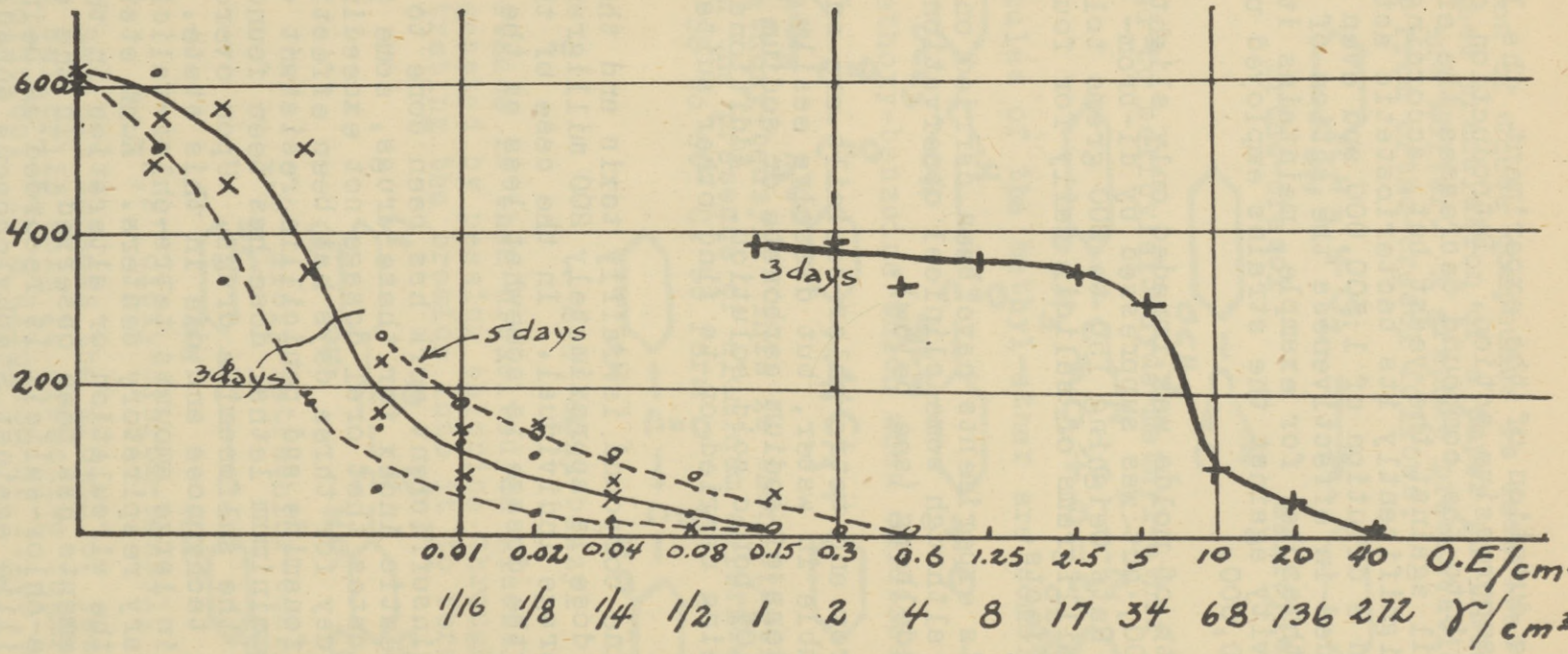
7. The growth of molds was impeded only slightly by salicil in 1:100, but was suppressed by di-brom-salicil, 1:1000. Rats weighing 150 to 200 grams tolerated oral doses of 50 milligrams of salicil daily for four days without unusual effects.

8. Dr. Kuhn's experiments have been carried on largely in vitro, although some clinical observation is in progress at Heidelberg (see below).

9. The di-brom and tetra-chlor-salicils are only very slightly soluble in water, but dissolve easily in very dilute solutions of sodium carbonate or sodium bicarbonate and in 10% boric acid solution. Additional experiments are being carried on to find other suitable solvents.

10. The bromine compound is fairly toxic and the maximum tolerated dose is approximately 200 milligrams per day for an average individual. In the case of the chlorine compound the dosage is somewhat less as the drug is more toxic.

11. Although insufficient work has been done to calculate a therapeutic index for these drugs, some preliminary work indicates that oral dosage not exceeding 20 milligrams per day for three days has been effective in clearing up sulfonamide and penicillin resistant cases of gonorrhea. The minimum lethal dose has been found to be the same as for the sulfonamide drugs. Upon overdosage, animals react with tachypnoea and die in this state. It is suspected that in large amounts tetra-chlor-salicil attacks the medullary respiratory centers. Kuhn states that no effect on the circulation or alteration in the essential blood elements has been observed. In the accompanying chart tetra-chlor-salicil is recorded as being as effective as penicillin against Staphylococcus aureus.


600
400
200
3 days
5 days
3 days
0.01 0.02 0.04 0.08 0.15 0.3 0.6 1.25 2.5 5 10 20 40 O.E/cm³
1/16 1/8 1/4 1/2 1 2 4 8 17 34 68 136 272 γ/cm³
x——x Penicillin Staph. aureus (K311)
+——+ " " albus (A Prag)
•----• Tetrachlorsalicil K311
o---o A Prag

This graph also indicates that this substance is six times as effective as penicillin against Staphylococcus albus. The ordinate is the degree of clouding (turbidity) of the culture, and the abscissa the units of penicillin and the quantities of tetra-chlor-salicil measured in gammas. The penicillin used in these experiments was captured by the Germans in the battle of the Ardennes Bulge in the winter of 1944. It consisted of tablets of the calcium salt of penicillin produced by Burroughs, Wellcome and Co., London. The lot number was 37-Ap-3012, dated September 1944.

12. The biochemical activity of these compounds is likewise being studied by Dr. Gerhard Domagk who will be found at the I.G. plant, Elberfeld, Strasse der S.A. 214. Dr. Helmuth Weese, at the same address, is doing the pharmacologic work. He has found the tetrachloride (3014) to be somewhat more toxic but no more effective than the dibromide (3065). Three thousand grams of the dibromide have been produced at Elberfeld but because of war conditions it has not been possible to deliver it to Kuhn at Heidelberg.

13. According to Kuhn, it is expected that further clinical experience with salicil compounds will demonstrate their therapeutic efficacy in those conditions in which penicillin is now the therapy of choice. He is attempting to establish salicil as an equally effective chemotherapeutic agent which has the advantages of being easy to manufacture at a very low cost and of being a versatile pharmaceutical which can be administered parenterally, orally, or topically.

14. It is not possible at present to say whether these expectations will be borne out. With the failure of the people at Elberfeld to deliver any large amounts of salicil, physicians carrying on clinical research have had to rely upon the small amounts prepared in the laboratories of the KWI by Kuhn and his associates. This has naturally limited the number of cases in which it has been used. Professor Hans Runge, of the University Gynecological Clinic (Universitäts Frauenklinik) stated that he had used the 3065 preparation in six women suffering from gonorrhea. Each of these patients had received four courses of sulfathiazole (Eleudron-"Bayer") without any permanent change in the smear. Dibromsalicil (3065) in the same amount caused the smears to become negative in all six cases. There was a remission in one woman whose smear became positive again after her next menstrual period. In these cases the drug was administered orally.

15. Professor Richard Siebeck stated that he had used

salicil in only one case. The condition was a chronic staphylococcal septicemia of four months duration. Repeated courses of sulfathiazole had apparently only maintained the status quo in the case. The patient exhibited a constant low grade fever which remained unaffected by sulfonamide administration. Repeated intramuscular injections of a bicarbonate solution of salicil rendered the patient afebrile within fourteen days and his progress since then has been steady. As there was no opportunity for Major Enloe to see this patient or to inspect the clinical case record, the available information is sufficient only to suggest the necessity of further investigation at the Heidelberg medical clinic.

16. Professor Bauer, director of the surgical clinic at Heidelberg demonstrated five (5) cases in which salicil has been applied locally. The first case was one in which the first and second fingers and the anterior third of the right hand had been amputated. The patient had received X-ray treatment for a very severe dermatitis in this area. The treatment was followed by severe X-ray burns and infection necessitating amputation. The wound was large and suppurative, with an extensive area of necrosis. Amputation had been performed 21 days before the visit had been made by Major Enloe. The wound had been treated only with finely powdered sterile salicil, and has been covered daily with dry dressing. When seen, the wound presented a healthy red appearance with granulation tissue throughout its entire area. Epithelialization was progressing around the edges. In general, the appearance of the patient, a man of middle age, was good.

17. The second patient was a German soldier who had been wounded by artillery fire, so that it had been necessary to amputate his right leg just above the knee. The wound was seven days old, and had been treated only by debridement without primary closure. Daily application of a thin layer of sterile salicil powder was stated to have produced healthy granulations with epithelialization. There was no evidence that the healing of the wound had been complicated by infection. The patient, a young man of about twenty-two, was cheerful, of good complexion, and is said to have had a good appetite after the first day.

18. The third patient had had both legs amputated in the middle third of the thigh. The appearance of the left leg was similar in all respects to that of the case described above (Patient 2). The surface of the right stump was marked

by a bone sequester near which was a deep tube abscess. The failure of this abscess to close like the remainder of the wound was attributed by Professor Bauer to the fact that it had not been possible to irrigate it with a salicil compound, nor because of the scarcity of the drug had it been possible to accompany local treatment with oral administration. Sulfonamides had not been given, Bauer continued, because he felt that the abscess would not alter the ultimate outcome of the case and he was reluctant to introduce any factor into this work which would becloud his appraisal of the therapeutic efficacy of salicil.

19. The fourth case was likewise a young German soldier whose right leg had been severed by artillery fire. This case was similar to that described above in paragraph 17, although epithelialization seemed to be progressing somewhat slowly.

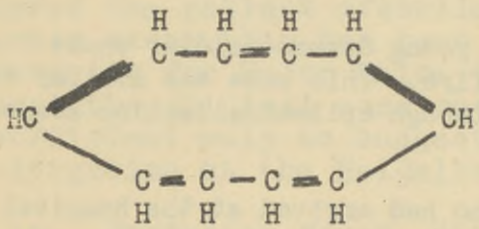
20. The fifth case was a patient who had arrived at the hospital 48 hours before being seen by Major Enloe. He had been struck in the right scapula by fragments of an artillery shell, and is reported to have lain for 36 hours before having been picked up. Upon arrival at the hospital the patient was found to have a massive suppurating wound. He was in a state of shock and running a high temperature. Professor Bauer said that he appeared somnolent in consequence of the absorption of wound toxins. Debridement necessitating the removal of three macerated muscle layers and fragments of bone and shell, was followed immediately by the application of a thin layer of sterile salicil. When seen, the patient appeared alert, cheerful, and of good complexion. The wound which had not been dressed since the operation, was partially covered by a layer of thick, yellow, purulent fluid; elsewhere small areas of healthy granulation tissue exuding clear serous fluid were noted. The dressing, upon removal, was saturated with both types of fluid. The wound was covered with salicil and dry dressings applied.

21. It was typical of each of these patients that they appeared quite healthy and showed no signs of intoxication. Upon questioning, each said that his appetite was good and that he felt well except for the wound itself. Each man appeared to be as alert and cheerful as could be expected in view of the psychological impact of their injuries. Closer examination of the wounds before fresh application of salicil did not reveal any caking of the powder which is so characteristic of the topical application of the sulfonamide drugs. Indeed, it appeared that the salicil was completely absorbed by the injured tissue and none of it could be found on the dressings or in the wound. Professor Bauer stated that in applying the powder the amount required was 1/10 by weight of that required of sulfonamides, in each case only an extremely thin layer of the yellow powder was applied.

22. Clinical trials carried out by other investigators in the

Kaiser Wilhelm Institut during the past few months have likewise been encouraging. Additional information on the clinical application of salicil compounds (blood levels, etc.) are being sought at present. In this connection, also, it is expected that pictorial records of the clinical effects will be made.

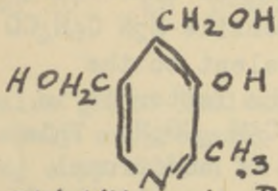
23. Cyclodekapentaene. Dr. Kuhn also reported the production of a substance called cyclodekapentaene, a cyclic hydrocarbon with the following structural formula:



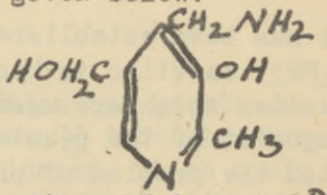
This product is a deep blue liquid having the approximate specific gravity of water. It is obtained as a by-product in the manufacture of butadiene (BUNA). The quantity of the by-product , cyclo-octa-tetraene, recoved in the manufacture of each 100 gallons of Buna is very small, but, Professor Kuhn stated, the amount of synthetic rubber produced at the Ludwigshafen plant of the I.G. Farben was sufficient to yield an almost unlimited amount of the eight-carbon molecule which was then acetylated with a catalyst. The ring formula given above can also be produced by the action of a strong catalyst on five molecules of acetylene. According to Kuhn, it has been found to be an extremely active agent against gram-negative bacilli. It is said to be very effective against most strains of B.coli, and has also been used successfully in experimental work as a decontaminant for mustard gas burns.

24. Alpha - tocopherol. In the course of experiments carried on at the Institute, Dr. Kuhn and his coworkers frequently observed that animals, fed on diets of highly purified protein, died. The protein used in these instances was casein which had been treated repeatedly to remove all traces of elements of the vitamin B complex. Autopsy of the animals almost invariably revealed fatty degeneration of the liver. Feeding of the experimental animals with casein was continued, but known amounts of alpha-tocopherol were added to the diet. It was observed that the addition of this factor was sufficient to prevent deleterious alteration in the fat metabolism of the liver. From these observations the inference was drawn that synthetic Vitamin E is closely associated with a fat - soluble reducing mechanism. Furthermore. It has led to clinical experimentation in the treatment of icterus, using 0.05 gm. doses of tocopherol twice daily. Professor Erich Grafe at Würzburg has reported successful results. On the other hand, Professor Hans Runge, director of the Frauenklinik at Heidelberg states that alpha-tocopherol has not been found to have any effect in eclampsia, which he believes to be due to faulty fat metabolism.

25. Vitamin B6. Professor Kuhn reported that when animals were fed on what is at present believed to be Vitamin B6, they exereted a substance which was more active than might have been expected from the quantities ingested. In other words, the animals appeared to be excreting more B6, than they had received; apparently, Vitamin B6 was being produced in the organism. The exereted substance was isolated and its structural formula determined. It is given below.



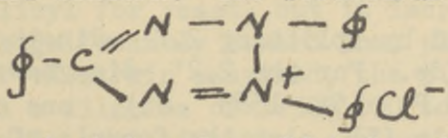
Old Vitamin B6



New Vitamin B6

On the basis of these findings the conclusion has been drawn that the substance excreted by the animals and isolated from the urine is actually Vitamin B6, while the substance hitherto regarded as such is in reality only a precursor or pro-vitamin.

26. Tetrazolium is a newly discovered agent which, by a very simple procedure, will provide a precise, quantitative fertility index for any given lot of seed. It was developed by Professor Kuhn in collaboration with Professor C. Lakon of the Bayer Company, and is being marketed by this concern. The structural formula of this substance is as follows:



ϕ is C_6H_5

Tetrazolium is synthesized from benzaldehyde, phenylhydrazine, and aniline. To obtain a fertility index for a given batch of seed, a 0.5 to 1.0% aqueous solution of tetrazolium is prepared. A sample (100 seeds) of the lot to be tested is then placed in this solution. For the most part, no preliminary treatment of the seeds is required; in testing some hard-shelled seeds, such as corn, the tips must be cut off so that the solution may penetrate. The number of seeds that turn red, as a result of the reducing action on tetrazolium of the active germinating substance contained within the seed, represents the percentage of fertile seeds to be expected in the lot from which the sample was taken. Late fermentation is also measured at the same time.

APPENDIX

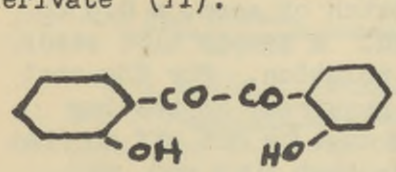
SALICIL

Kuhn, R., Birkofer, L., Müller, E. F. (authors).
(in: Berichte der Deutschen Chemischen Gesellschaft 76: 900-904, 1943)

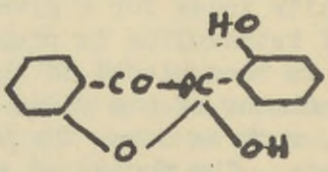
It has been established that 4.4' diamino-benzile $H_2N\ C_6H_4CO\ CO.C_6H_4NH_2$ in vitro is bacteriostatically equivalent to the sulfonamides which are used clinically, and as the latter it is an antagonist of the p-amino-benzoic acid, $H_2N\ C_6H_4CO_2H$. This initiated the problem of preparing the diketones of additional physiologically effective carbo-acids and of examining them biologically.

Frequently attempts have been made to prepare 2.2' -dioxy-benzile. (Salicil), corresponding to salicylic acid, but all these efforts have been in vain. It was possible to dehydrate salicylaldehyde to the osazone of 2.2' -dioxy-benzile, but neither the α-osazone thus obtained, nor the β-osazone obtained by boiling the α-osazone with nitrobenzile, could be split to form the desired salicyl. Also the present author's attempts along this line have failed.

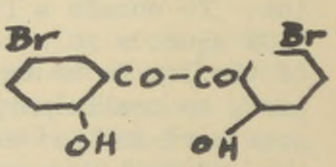
Results were obtained by splitting 2.2' -dimethoxy-benzile by means of aluminium chloride. For the 2.2' -dioxybenzile thus obtained (of yellow hue; M.P. 154 -158° C.), one must consider, besides the diketon formula (I), also the formula of a cumaranon derivate (II).



I



II



III

The reaction with phenylhydrazine in 70% acetic acid resulted in an osazone (m.p. 265 - 267°),. crystallizing in yellow platelets from nitrobenzol, which, when mixed with a control preparation obtained by transposition of the α-osazone according to H. Blitz, melted at 264 - 265°. With acetic acid anhydride the 2.2' - dioxy-benzile forms a white diacetyl compound (m.p. 123°), and with bromine the bright-yellow 5.5' dibrome 2.2'-dioxy-benzile (m.p. 213°). Proof for the position of the bromine atoms is rendered in the experimental section by the synthesis 5-brome-2-methoxy-benzaldehyde KON→ 5.5'-dibrome 2.2'-dimethoxy-benzoin (m.p. 105°) Fehling→ 5.5' -dibrome- 2.2'-dimethoxy-benzile (m.p. 232°) AlCl3→ 5.5'-dibrome 2.2'-dioxy-benzile (mixed tests).

Results of the tests on yeast (strain M), lactic acid bacteria (Sbm. plantarum 10S I.G.) and Staphylococcus pygenes aureus (strain

vR) were as follows:

(See chart p.901, in original)

*) for 10^{-9} gm p-amino-benzoic acid/cc
**) for 10 gm p-amino-benzoic acid/cc

The value gm/cc indicated at which concentration the growth was completely suppressed; > indicated incomplete, >> indicates totally wanting suppression at the concentration indicated, which could not be raised because of low solubility. The concentrations were arranged, as is customary, in powers of two.

The following are especially remarkable: a) in contrast to salicylic acid, salicyl impedes the growth of yeast independantly from the pH. At a pH= 6.6 it still suppresses the growth of the yeast examined completely at a dilution of 1:100 000; thus it is one of the most effective substances known. In the case of Staphylococcus, salicyl is less effective; especially with longer duration of the experiment the impeding action is greatly lowered. (b) Introduction of 2 bromine atoms into the 5.5' position lowers the efficacy of salicyl for yeast, but it increases the bacteriostatic activity significantly toward Sbm. plant. and Staph. aureus. In case of vR-strain examined, the 5.5' -dibrome- 2.2' -dioxy-benzile with a dilution of 1:500 000 to 1:1000 000 surpasses the sulfonamide most effective against this strain, namely sulfathiazole (1: 30 000) by about one power of ten, and even in repeated experiments with increasing duration of the experiment no decrease in its efficacy was determined.

For the same conditions, the following values were obtained with various sulfonamides for 2 strains of Staphylococcus pygenes aureus (E.F. Möller):

(See chart p. 902, of the original)

Salicyl 1:1000 had no suppressing effect upon alcoholic fermentation (compressed yeast and fruit yeast). The growth of molds (Penicillium glaucum, Aspergillus clavatus) was impeded but slightly by salicyl 1:100; it was, however, suppressed completely by means of 5.5'-dibrome-salicyl 1:1000 (nutrient solution according to Czapek-Dox). Rats weighing 150-200 gm tolerated 50 mg salicyl in olive oil perorally and subcutaneously daily for an experimental series lasting 4 days, without any unusual effects. Salicyl suppresses the sprouting of water-cress seeds (Lepidium sativum) even in a dilution of 1:10 000 (F. Moewus).

Description of the experiments.

2.2'-dioxy-benzile: 10 gm 2.2'-diomethoxy-benzile (m.p. 154° to 155°) were dissolved in 500 cc nitrobenzol and heated with 100 gm finely powdered aluminum chloride to 55° in an oilbath for 7 hrs. The mixture was then poured on ice, centrifuged, the aqueous layer was syphoned off and the nitrobenzol solution was then stirred energetically with an equal volume of 2n NaOH solution for 30 min. The alkaline solution, separated by the centrifuging, was deep yellow; when it was rendered acidic by the addition of 2n Sulfuric acid, it furnished 5.0 gm 2.2'-dioxy-benzile (m.p. 150-152°), i.e. 50 - 60% of the theoretical output. By recrystallization with 70% acetic acid one obtains coarse needles of yellowish hue (m.p. 154 - 155°).

3.790 mg substance: 9.655 mg CO_2, 1.46 mg H_2O. -- 0.411 mg substance in 4.43 mg exalton: Δ - 0.85°.
$C_{14}H_{10}O_4$, Calc.: C 69.40, H 4.16, Mol. Wt. 242 Found: C 69.48, H 4.31 Mol. Wt. 236

In melting, the 2.2'-dioxy-benzile becomes deep yellow. With cooling, the coloration again becomes lighter. The Alcoholic solution with ferric chloride has a brownish coloration. Solubility is very slight with water and bicarbonate solution. In dilute soda solution and dilute NaOH solution the substance dissolves readily, with deep yellow coloration. In alcohol the color is light yellow, in benzol it is deeper.

Analysis of osazone (m.p. 265 - 267°).

$C_{26}H_{22}O_2N_4$ (422.2), Calc.: C 73.90, H 5.25, N 13.27; found: C 73.62, H 5.26, N 13.1 .

The diacetyl compound obtained by boiling with acetic acid anhydride for 30 min. crystallizes from alcohol in white scales of m.p. 123°. $C_{18}H_{14}O_6$ (325.1) Calcul. : C 66. 24, H 4.33 Found: C. 66.57, H 4.55

5.5'-dibrome-2.2'-dioxy-benzile: With bromine in glacial acetic acid the 2.2'-dioxy-benzile produces a dibrome compound of brightly yellow coloration, which one obtains from glacial acetic acid in flexible needles of a m.p. of 212 - 213°. In melting, the coloration becomes deeper, toward orange; when cooled, the color again becomes lighter.

5.5'-dibrome 2.2'-dimethozy-benzoin: 10 gm 5-brome-2-methoxy-benzaldehyde were dissolved in 50 cc 50% alcohol and boiled with 1 gm potassium cyanide for three hours with a reflux condenser. Then the mixture was poured with energetic stirring into a beaker cooled by means of a salt-ice mixture; the very viscous mass was caused to crystallize by rubbing with 5 gm of absolute alcohol. For analysis the substance was recrystallized from a small quantity of absolute alcohol. Pale, coarse platelets of yellow hue, m.p. 105°. $C_{16}H_{14}O_4Br_2$ (429.9) Calcul. C 44.66 H 3.28, Br 37.18. Found: C 44.59, H 3.47, Br 36.94.

5.5'-dibrome-2.2'-dimethoxy-benzile: 5 gm dibrome-dimethoxy-benzoin were dehydrated in 70 cc 70% alcohol with exactly the required amount of Fehling's solution at boiling heat (reflux). By boiling the Cu_2O ppt. and shaking the supernatant solution with chloroform the authors obtained 4.5 gm 5.5'-dibrome 2.2'-dimethoxy-benzile. Crystallization from glacial acetic acid or alcohol produced coarse white needles. m.p. 232°.

$C_{16}H_{12}O_4Br_2$ (427.9) Calcul.: C 44.87, H 2.83. Found: C 45.04, H 3.03.

The same compound was obtained from 2.2'-dimethoxy-benzile and bromine (2 mol.) in glacial acetic acid. m.p. 232 - 233°. Mixed m.p. 231-233°.

5.5'-dibrome-2.2'-dioxy-benzile: 1 gm dibrome-dimethoxy-benzile was heated in 50 cc nitrobenzol with 10 gm aluminum chloride for 7 hrs. to 55° in an oil bath. The experiment (cf. 2.2'-dioxy-benzile) yielded 0.52. gm. 5.5'-dibrome-2.2'-dioxybenzile (56% of the theoretical output), which crystallized from glacial acetic acid in flexibile, lemon-yellow needles, m.p. 210°.

$C_{14}H_8O_4Br_2$ (399.9) Calcul.: c 42.01, H 2.03. Found: C 42.12, H 2.33.

The same compound was obtained by the authors from 2.2'-dioxy-benzile by bromination in glacial acetic acid. m.p. 212-213°; mixed m.p. 212°. Readily soluble in alcohol and benzol, soluble in ether, almost insoluble in water; it is absorbed by dilute NaOH-solution with deep yellow coloration. The boric acid complex of 5.5'-dibrome-salicyl is readily soluble in water, its solutions are colorless in the cold and become yellow when heated. The boric acid complex is equally effective against Staph. pyogenes aureus Vr as the free compound (full suppression of growth by 1 - 2 x 10^{-6} gm/cc). For the bacteriological tests, 5 mg dibrome-dioxy benzile-6 mg $Na_2B_4O_7$ - 10 H_2O were dissolved in 3 cc of water; the p.H. was raised to 7.5 by means of 0.1 cc m/15 KH_2PO_4' and distilled water was added to raise the total volume to 5 cc. 50 mg 5.5'-dibrome-salicyl require approx. 5000 cc pure water, but they dissolve clearly in 1 cc if one adds 31 mg H_3BO_3 20 mg NaOH and finally adds 25 mg KH_2PO_4 as a buffer.

2-oxy-2'-methoxy-benzile: 2.2'-dimethoxy-benzile (2gm), by boiling in 20 cc glacial acid with 5 cc 66% hydrobromic acid for 10-20 min, yielded, besides other bromine-containing compounds and unchaged original material by frequent recrystillization from absolute alcohol (considerable cooling required, envelope-like scales, m.p. 124 - 126°, representing the bromine-free phenol fraction. $C_{15}H_{12}O_4$ (256.1) Calcul.: C 70.29, H 4.72. OCH_3 12.10.

Found C 70.01, H 4.54, OCH_3 12.14.

The melted substance is deep yellow and becomes again colorless upon cooling.