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HEXYL RESORCINOL

THE DEVELOPMENT AND CLINICAL APPLICATION OF A SYNTHETIC COMPOUND POSSESSING THE EXPERIMENTAL REQUIREMENTS OF AN IDEAL INTERNAL URINARY ANTISEPTIC

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The majority of those confronted with the necessity of treating infections of the urinary tract are quite willing to acknowledge that this field of therapy is one which admits of very wide improvement. The brilliant advances in urologic diagnosis made possible by the various technical procedures now at our command have not been accompanied by equally satisfactory results in the treatment of infections of the urinary apparatus in spite of the fact that they have made every portion of its mucosa accessible to direct treatment with bactericidal substances.

This fact is readily explained.

In general there can be but two direct methods of controlling a urinary infection: First, the application of bactericidal substances to the mucosa by topical application of lavage, and secondly, the internal administration of some substance calculated to impart to the urine either bacteriostatic or bactericidal properties. Since no drug supposed to possess either of these virtues has been found by general clinical use permanently to influence the course of chronic urinary tract infections, it has been necessary to depend upon the first method in spite of the many serious objections and obstacles which limit its employment.

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THE FUTURE OF INTERNAL ANTISEPSIS IN ITS APPLICATION TO UROLOGY

We need only witness the difficulty experienced in disinfecting inflamed mucous surfaces far more accessible to treatment than the urinary tract, such as chronic infections of the naso-pharynx for instance, or the gonorrheal vaginitis of little girls to realize that any great therapeutic expectations based on the direct intermittent application of bactericidal substances to the infected mucosa must be materially discounted, however powerful a germicide should become available. That the tissue reaction initiated by the irritant properties of the substance employed is of importance in effecting a biological rather than a direct chemical disinfection cannot be denied, but in order to explain the high percentage of cures in the treatment of such conditions as chronic pyelitis not infrequently reported, even after making full allowance for those cases which have been "repeatedly cured" only to become promptly reinfected from some distant focus, it is necessary to assume that this tissue reaction, which is nothing more nor less than a chemical inflammation, plays a far more important rôle in the urinary tract than elsewhere. Whatever the explanation, it seems highly probable that any large advances in the treatment of chronic infections of the urinary tract must come from investigation of the possibilities of internal antisepsis.

The internal administration of a substance which would yield favorable clinical results in urinary infections even approaching those obtained by any method necessitating instrumentation, would mark a very great advance. Even if renal lavage with some newly discovered germicide were found to be completely effective in every case of pyelitis, any non-toxic drug which, by simple internal administration would effect a cure in only a small percentage of these cases, would be clearly indicated in most instances before proceeding to the more radical measures. Moreover, an internal urinary antiseptic which could not be counted upon to do more than reduce the number of instrumentations necessary to a cure would be highly acceptable. Were any drug available which could be trusted to perform any of these functions

with regularity it would now be universally employed. It is not surprising therefore that the importance of this problem has stimulated a great deal of splendid scientific work.

By far the most comprehensive investigation of this problem has been conducted for several years past at the Brady Urological Clinic of the Johns Hopkins Hospital by Dr. Hugh H. Young and his associates (1, 2, 3, 4, 5, 6). We are indebted to these investigators for a clear and concise statement of the experimental qualifications necessary to the ideal urinary antiseptic: It should be chemically stable, non-toxic and non-irritating to the urinary tract; it should exert an antiseptic action in high dilution in urine of any reaction and should be eliminated in high percentage by the kidney. After a study of over 400 compounds many of which were synthesized for the purpose, Davis concluded that no such substance had ever been described. As late as March, 1921, he states (7): "There is no sound experimental or clinical proof of the fitness of any known drug for use as an internal urinary antiseptic."

The problem as a whole is more complicated than would appear at first sight. We know that these qualifications are essential to an internal urinary antiseptic in the usual sense of the term, but for the reason that we have not had the opportunity of proving it, we do not know that any substance possessing them would necessarily be clinically successful in all types of infections of the urinary apparatus. For certain very definite reasons which will be mentioned it is even to be expected that such a substance would not be uniformly successful. There will be described in this paper a new substance—hexyl resorcinol—which possesses all of the qualifications mentioned, with the added advantages that its action in the urine is bactericidal rather than "antiseptic" and that inasmuch as it is administrable by mouth in repeated doses for indefinite periods, an almost continuous, rather than an intermittent action in the urinary tract is readily obtainable. Yet this substance has its failures in spite of its ideal experimental qualifications and in spite of the fact that it has yielded remarkable clinical results in many types of urinary infections. These may be due in part to actual failure of the drug

itself in some instances and in part to certain limitations which must necessarily qualify the clinical value of any urinary antiseptic, however closely it approaches the commonly accepted experimental ideal.

Depending, as we must, upon the secretion of the active substance in the urine, a functioning kidney is of course the first necessity. Furthermore no urinary antiseptic could be expected to exert any influence on infection located in an area with which the urine is not in intimate contact. This at once limits the field of urinary antisepsis to the surface of the mucosa of the urinary tract. If the infection has penetrated beneath the mucosa it would be unreasonable to expect prompt disinfection by any urinary antiseptic although it is conceivable that it might exert a favorable influence upon its course. If, on the other hand, the infection has invaded the parenchyma of the kidney, we are confronted with even a greater difficulty.

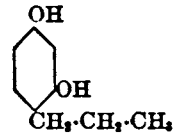
Secretory affinities and tissue affinities are very different physiological phenomena. For certain reasons it is quite improbable that any substance possessing Davis' qualifications, which include its secretion into the urine in high percentage, would disinfect the renal parenchyma during its passage from the blood stream into the urine, although, again, it is conceivable that the course of an infection lying in its path might be favorably influenced. To accomplish disinfection of the kidney tissue itself it would be necessary that such a substance either retain its bactericidal action in the blood stream or at least during its passage through the renal cells by which the process of its excretion is controlled. A substance retaining its bactericidal action in the blood stream would probably qualify as a general systemic disinfectant and would possess far more important clinical applications than that of an internal urinary antiseptic, while even a substance which merely became active coincident with its passage through the renal epithelium might naturally be expected to disinfect that tissue to better advantage if it were retained or bound rather than if it were rapidly excreted by it. It would seem, therefore, that an internal antiseptic calculated to disinfect the kidney tissue itself should possess experimental

qualifications quite different from those essential to a drug intended to disinfect the urinary mucosa by imparting bactericidal or bacteriostatic properties to the urine. Experimental evidence to be reported in a future communication points strongly to the assumption that *internal renal antisepsis* is a problem essentially different from *internal urinary antisepsis*, that an internal renal antiseptic must be *bound* by renal tissue while an internal urinary antiseptic must be rapidly *excreted* by it. It is difficult to conceive of any substance possessing both these qualifications and it seems highly improbable that any one substance would satisfy perfectly either the experimental or clinical demands of both these fields.

It would appear that efforts to discover a more efficient germicide intended for direct application to the urinary mucosa, hold insufficient promise to justify the work involved, not only because of the limitations inherent in this method of disinfecting mucous surfaces in general and the technical difficulties involved in its employment in this particular field, but chiefly because even if such a substance were successfully developed it would still result in no great improvement on our therapy of infections of the urinary tract as a whole. On the other hand, in spite of the complications and difficulties peculiar to the problem of internal antisepsis in its relation to urology, any genuine advance in this field offers every prospect of decided improvement in our clinical results.

FACTORS LEADING TO THE SYNTHESIS OF HEXYL RESORCINOL AND ITS EXAMINATION AS AN INTERNAL URINARY ANTISEPTIC

The synthesis of the lower homologues of a series of alkyl resorcinols by Johnson and Hodge (8, 9) in 1913 was the starting point of this investigation. At this time Rettger (8, 9) found there was a pronounced increase in bactericidal properties with the addition of each carbon atom to the alkyl chain, while a preliminary biological examination by the writer revealed the fact that there was a coincident diminution in toxicity to laboratory animals, so pronounced that in n-propyl resorcinol,



there existed a relatively non-toxic substance with several times the bactericidal power of phenol. Furthermore it was found that this substance was excreted in the urine after oral administration.

TABLE 1

*Phenol coefficients of the alkyl resorcinols and some well known germicides**

* Resorcinol	0.3	Cresol saponified	1.03
* Ethyl resorcinol	1.50	Carbolene	1.36
n-Propyl resorcinol	5.00	Lysol	2.12
Iso-butyl resorcinol	15.20	Trikresol	2.62
n-Butyl resorcinol	22.10	Crude carbolic acid	2.75
Iso-amyl resorcinol	23.80	Cresoleum	2.90
n-Amyl resorcinol	33.00	Liquor cresolis comp. (U. S. P.)	3.00
Iso-hexyl resorcinol	27.30	Creolin (Pearson)	3.25
n-Hexyl resorcinol	46.00		
n-Heptyl resorcinol	30.00		

* The phenol coefficients of the commercial germicides listed are those recorded by McClintic, Hygienic Laboratory Bulletin No. 82, April, 1912, p. 35. Those of resorcinol and ethyl resorcinol are taken from the article by Johnson and Lane, Jour. Amer. Chem. Soc., 1921, p. 348. I am indebted to Dr. J. C. Swenarton for the remaining determinations which were made in accordance with the standard requirements of the Hygienic Laboratory method now in use as described in Pub. Health Reports, July 8, 1921.

Having established the fact that, up to this point at least, alkylation of resorcinol resulted in an increase in bactericidal power and a decrease in toxicity directly proportional to the molecular weight of the fatty chain, it became desirable to determine just how far these effects could be carried. Application of this principle to practically every available phenolic nucleus yielded indifferent results which will not be reported at this time. With resorcinol as the mother substance, however, a series of compounds was obtained which must be numbered among the most powerful organic germicides ever described. An idea of their activity as compared with a number of the ordinary poisonous

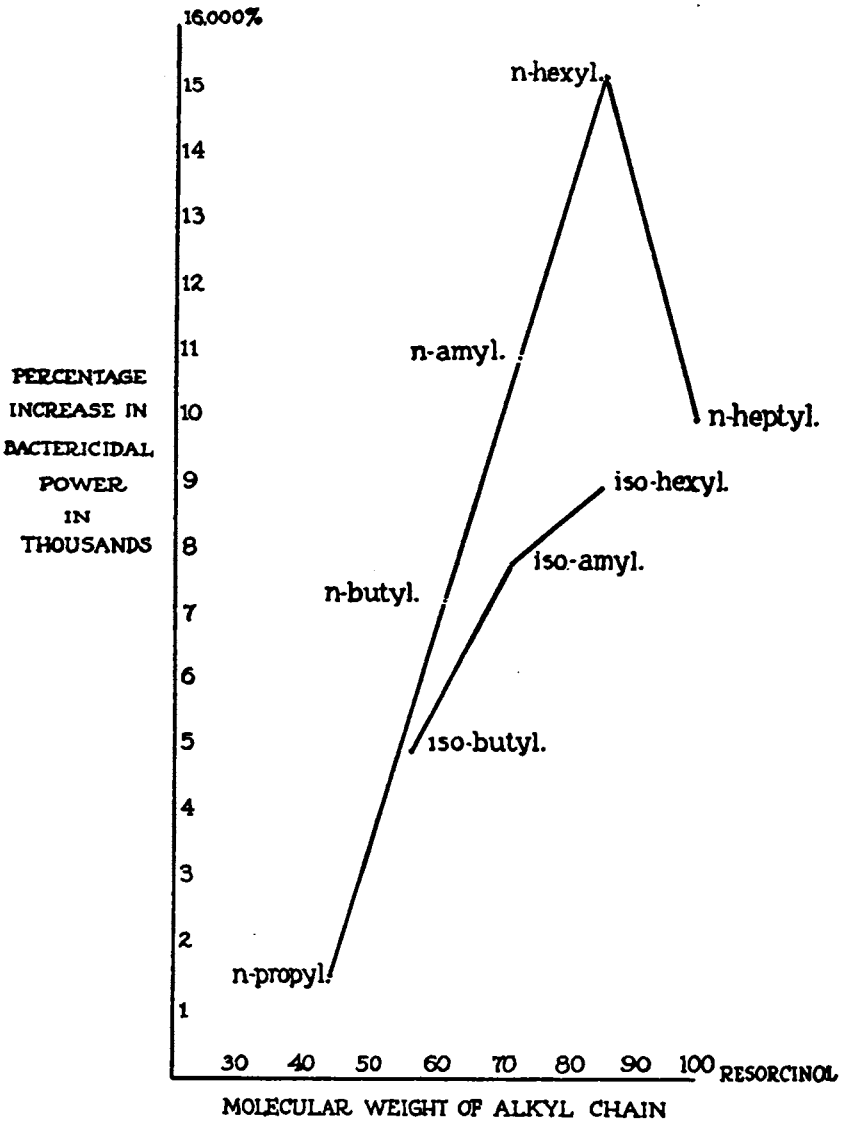
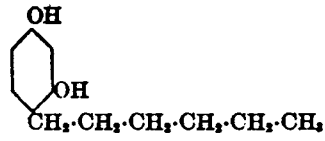


FIG. 1. ALKYL RESORCINOLS

The increase in bactericidal power over the mother substance (expressed in thousands of per cent) is directly proportional to the molecular weight of the alkyl chain. The experimental points for the normal derivatives may be regarded as lying upon a straight line. The variations are probably within the limit of error.

germicides in common use may be had by consulting table 1. As shown in figure 1, the peak of bactericidal power is reached in n-hexyl resorcinol,



which, with a phenol coefficient of 46, exhibits an increase in bactericidal power over resorcinol or more than fifteen thousand per cent. It is by far the most powerful germicide ever described as a non-toxic substance. Like its homologues it is excreted in the urine, after oral administration to rabbits and to man and imparts to the urine active bactericidal properties.

The secretion of the higher alkyl resorcinols in the urine after oral administration to rabbits was discovered in an interesting manner. Solutions of propyl resorcinol yield a red brown color on the addition of a few drops of ferric chloride, as does also the urine of rabbits fed on this substance. With the addition of the fourth carbon atom in the alkyl chain in the butyl derivatives, however, this color reaction is lost. While attempting to determine the presence of iso-butyl resorcinol in the urine of a rabbit which has received a large dose of this substance by mouth, by destroying the alkyl chain and then applying the usual test for resorcinol, concentrated nitric acid was added. A brilliant red color appeared at once. All of the alkyl resorcinols give this test[2] in aqueous solution and their presence in rabbit urine may be readily determined by means of it. Gentle heat is necesaary to bring out the red color with propyl resorcinol; the higher homologues give it in the cold; iso-hexyl and n-heptyl resorcinol yield an orange color.

Having determined that each of the alkyl resorcinols from the propyl derivative up was secreted into the urine after oral

[2] This test, which has not been previously described, is given by resorcinol itself, gentle heat being necessary to bring out the color. It is more than ten times as delicate as the usual ferric chloride test for this substance. Orcin and phloroglucinol also yield a red color.

administration to rabbits, it remained to be determined which of these substances showed the greater promise of usefulness as an internal urinary antiseptic.

Although bactericidal urine was obtained occasionally in rabbits after oral administration of each of the alkyl resorcinols mentioned, the hexyl and heptyl derivatives proved to be very much superior to the lower homologues in this regard. Of these two, hexyl resorcinol was found to be the more promising and detailed discussion will therefore be limited to it. For the reason, however, that heptyl resorcinol meets all the experimental qualifications, though less decisively, it will be contrasted with hexyl resorcinol as opportunity permits.

TABLE 2

Bactericidal properties of higher alkyl resorcinols, showing maximum dilutions destroying test organisms in water and in human urine

DERIVATIVE	TEST ORGANISM	WATER		ACID URINE pH 6.0 TO pH 6.4		ALKALINE URINE pH 7.6 TO pH 8.2	
		One hour	Twenty-four hours	One hour	Twenty-four hours	One hour	Twenty-four hours
Hexyl resorcinol	B. coli	12,000	30,000	10,000	10,000	10,000	18,000
	Staphylococcus albus	100,000	110,000	60,000	70,000	18,000	60,000
Heptyl resorcinol	B. coli	10,000	30,000	6,000	6,000	10,000	10,000
	Staphylococcus albus	60,000	70,000	50.000	90,000	40,000	80,000

EXPERIMENTAL QUALIFICATIONS OF HEXYL RESORCINOL AS AN INTERNAL URINARY ANTISEPTIC

1. *Hexyl resorcinol is a stable chemical compound* withstanding repeated distillation under diminished pressure. The same is true of heptyl resorcinol. Both are white solids of a somewhat waxy nature.

2. *Hexyl resorcinol is bactericidal in high dilution in urine of any reaction.* Heptyl resorcinol also meets this requirement. The technic by which the results recorded in table 2 were obtained has been previously described.[3]

[3] In press. Jour. Amer. Med. Assoc.

3. Administered by mouth, hexyl resorcinol is non-toxic to rabbits and to man. Not only do rabbits tolerate oral administration of very large doses (1 to 2.5 grams) of hexyl resorcinol and its homologues, but remain normal in all respects when fed large daily doses (0.5 gram) for a period of two or three weeks. Following these intensive courses the animals kept for observation over a period of fourteen months have maintained their weight and the urine and kidney function have remained normal, while the organs of those animals sacrificed at the end of the course were found to be normal on gross and microscopic examination. See table 3.

During a period of ten weeks, hexyl resorcinol and heptyl resorcinol were administered to five normal men in doses ranging from 0.15 to 1 gram three times a day. Each subject received a total of over 70 grams. In no instance was there any evidence of any toxic effect nor has there been during the eight months since the conclusion of the experiments. A number of patients with urinary tract infections have since consumed hexyl resorcinol in much larger quantities than this without any toxic effect whatever.

The first few doses of hexyl resorcinol may cause some intestinal irritation as evidenced by griping and catharsis. Two of the five normal subjects experienced this. These effects disappear promptly in all but very exceptional instances, even though the dosage is increased. There may be a definite diuretic effect in both rabbits and man following the first few doses of either hexyl or heptyl resorcinol. Diuresis, like catharsis, ordinarily disappears, however, even though the dosage is increased.

Were it not for the fact that hexyl resorcinol forms a very soluble, non-irritating salt in the alkaline intestinal juices, it might be too irritating for clinical use. The sodium salt cannot be administered directly because the free substance would be precipitated in the stomach. Hexyl resorcinol has therefore been given in capsules or pills coated with salol to prevent or retard disintegration in the stomach. If taken after meals gastric irritation is very rare.

4. Hexyl resorcinol by mouth is non-irritating to the urinary tract. During intensive courses of hexyl and heptyl resorcinol administered to rabbits and to normal men, a total of 140 specimens of

TABLE 3

Prolonged administration of the higher alkyl resorcinols to rabbits

	DERIVATIVE			
	Normal hexyl resorcinol		Normal heptyl resorcinol	
	Rabbit 5	Rabbit 22	Rabbit 30	Rabbit 27
Daily dose	0.5 gram (0.222 gram p.k.)	0.5 gram (0.198 gram. p.k.)	0.5 gram (0.166 gram p.k.)	0.5 gram (0.2 gram p.k.)
Administered for	21 consecutive days	21 consecutive days	10 consecutive days	10 consecutive days
Total given	10.5 grams (4.66 grams p.k.)	10.5 grams (4.55 grams p.k.)	5 grams (1.66 grams p.k.)	5 grams (2 grams p.k.)
Equivalent dosage for 150 pound man	305 grams	298 grams	108 grams	131 grams
Weight at start	2250 grams (August 7)	2530 grams (August 7)	3000 grams (August 20)	2500 grams (August 20)
Urine at start	Normal	Normal	Normal	Normal
Kidney function	80 per cent (phenolsulphonephthalein)	70 per cent (phenolsulphonephthalein)	70 per cent (phenolsulphonephthalein)	70 per cent (phenolsulphonephthalein)
Fate	Saved for observation	Sacrificed August 28	Saved for observation	Sacrificed August 30
Subsequent observations and autopsy findings	Weight August 28 2350 grams Weight September 28 2675 grams Weight October 28 3100 grams Weight December 3 3375 grams Weight December 27 3425 grams Phenolsulphonephthalein August 28 80 per cent Phenolsulphonephthalein September 28 70 per cent Phenolsulphonephthalein October 28 80 per cent Phenolsulphonephthalein December 3 80 per cent Phenolsulphonephthalein December 27 70 per cent Urine August 28 normal Urine September 28 normal Urine October 28 normal Urine December 3 normal Urine December 27 normal	Weight August 28 2450 grams Phensolsulphonephthalein August 28 70 per cent Urine August 28 normal *Autopsy* All tissues normal in gross appearance. Following tissues found to be normal on microscopic examination Kidney Liver Spleen Pancreas Adrenal Stomach Duodenum Ileum Caecum Rectum Ureter Bladder Heart Lung Optic nerve	Weight August 28 3075 grams Weight September 28 3250 grams Weight October 28 3600 grams Weight December 3 3850 grams Weight December 27 3900 grams Phenolsulphonephthalein August 28 70 per cent Phenolsulphonephthalein September 28 70 per cent Phenolsulphonephthalein October 28 70 per cent Phenolsulphonephthalein December 3 70 per cent Phenolsulphonephthalein December 27 70 per cent Urine August 28 normal Urine September 28 normal Urine October 28 normal Urine December 3 normal Urine December 27 normal	Weight August 30 2550 grams Phenolsulphonephthalein August 30 80 per cent Urine August 30 normal *Autopsy* All tissues normal in gross appearance Following tissues found to be normal on microscopic examination Kidney Liver Spleen Pancreas Adrenal Stomach Duodenum Ileum Caecum Rectum Ureter Heart Lung Optic nerve

rabbit urine obtained by catheter and 826 specimens of urine from the five human subjects were examined for albumin, pus, blood and casts. In no instance was any abnormality discovered. Furthermore the functional activity of the kidneys, which was determined to be normal in both rabbits and men before the course was begun, remained normal during the course and for several months of observation following its completion.

In rabbits the administration of large doses is followed by the appearance of conjugates in the urine which interfere with the usual tests for albumin. No trace of albumin could be found in any of the specimens with the potassium iodide test (10), which will detect the presence of albumin in concentrations as low as 0.005 per cent. In man there is no interference with the usual tests for albumin.

5. Hexyl resorcinol is eliminated in high percentage by the kidney —the larger portion as a conjugate. Following relatively large single doses of hexyl resorcinol to rabbits, the urine secreted by these animals within one hour and not infrequently as long as twenty-four hours after the dose, will show a red color on the addition of concentrated nitric acid. This test is positive in human urine only after very large doses. The fact soon developed that the bactericidal properties of a given specimen of rabbit urine could not be forecasted by the concentration of substances in it which produce a red color with nitric acid. Many specimens showing a brilliant red color with this test were found to be devoid of bactericidal properties, while a specimen giving a very faint test or none at all was not infrequently found to be actively bactericidal. Furthermore, rabbits were found sometimes to secrete bactericidal urine intermittently for twenty-four hours after large single doses and entirely without any relation to the depth of the red color produced in these specimens by nitric acid. Davis and Hain (11) have shown that both rabbits and dogs may normally secrete bacteriostatic urine. The experiments were of course carefully controlled to avoid misinterpretation in this regard, despite the fact that so far as is known bactericidal urine is not secreted by these animals.

The conclusion that the bulk of each dose of hexyl resorcinol is eliminated as a conjugate which has no bactericidal action but which yields (in rabbits) a red color with nitric acid, has much to support it. The secretion of bactericidal urine probably depends upon the secretion of free hexyl resorcinol and there is abundant evidence that the amount is never more than a very small percentage of the quantity ingested. The fate of this substance in the body is now being investigated.

TABLE 4

Secretion of bactericidal urine by rabbits

INTERVAL SINCE LAST DOSE	NUMBER OF URINE SPECIMENS	BOTH ORGANISMS KILLED	B. COLI (ONLY) KILLED	STAPHYLOCOCCUS ALBUS (ONLY) KILLED	NEITHER ORGANISM KILLED	NUMBER OF BACTERICIDAL URINES	PERCENTAGE OF BACTERICIDAL URINES
Normal hexyl resorcinol; 6 rabbits; doses, 0.2 to 0.5 gram.							
Hours							*per cent*
1	17	7	0	2	8	9	53
2	4	1	0	2	1	3	75
3	13	6	0	6	1	12	92
4	11	6	0	2	3	8	73
5	10	3	0	5	2	8	80
6–8	6	4	0	0	2	4	66
18–28	18	6	0	4	8	10	56
Totals...	79	33	0	21	25	54	68
Normal heptyl resorcinol; 4 rabbits; doses, 0.1 to 0.5 gram							
½–1	14	5	4	0	5	9	64
3	4	1	0	0	3	1	25
4–6	10	2	1	0	7	3	30
24	17	1	2	0	14	3	17
Totals...	45	9	7	0	29	16	35

SECRETION OF BACTERICIDAL URINE BY RABBITS

Following oral administration of single doses of hexyl and heptyl resorcinol to rabbits, the urine secreted by these animals within one hour and as long as twenty-four hours after, frequently possesses bactericidal properties. The details of these experiments are summarized in table 4. The technic employed has

been previously described[4] and will not be repeated except to say that 1 cc. amounts of these urine specimens were each innoculated with one 3 mm. standard loopful of a twenty-four hour broth culture of strains of B. coli and Staphylococcus albus isolated from urinary infections and then incubated for eighteen to twenty-four hours. If one or both organisms were found to have been completely destroyed, the specimen was recorded as bactericidal. If less than 10 colonies appeared on the transfer the result was recorded as scant, but all such specimens were recorded as devoid of bactericidal properties in summarizing

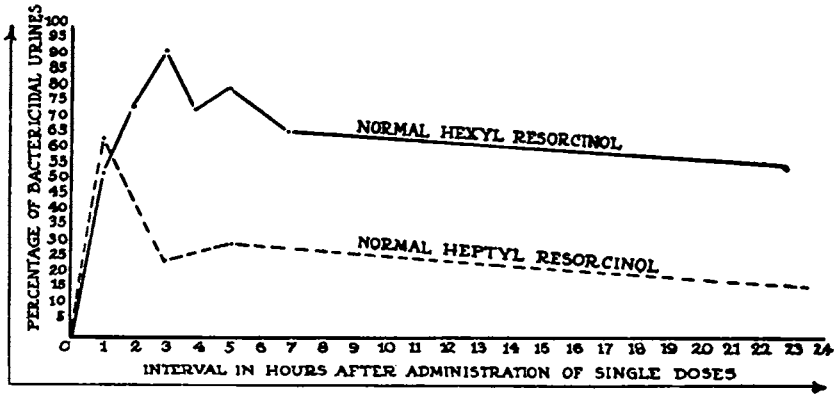


FIG. 2. RELATIVE EXPECTANCY OF BACTERICIDAL URINE IN RABBITS
At various intervals after single doses of n-hexyl n-heptyl resorcinol

the results. Many of the specimens recorded as inert were found to be bactericidal to one or both organisms if 5 cc. portions were used in the tests instead of 1 cc. portions. While these results will not be reported in detail in this paper, it may be said that mass action plays an important rôle in these experiments and helps to explain certain of the clinical results.

The percentage of bactericidal urine specimens obtained at various intervals after single doses of hexyl and heptyl resorcinol is given in table 4. The superiority of hexyl resorcinol over the heptyl derivative is made even more evident in figure 2, which shows the relative expectancy of bactericidal urine in rabbits at intervals after single doses of these substances.

[4] In press. Jour. Amer. Med. Assoc.

TABLE 5

Secretion of bactericidal urine by man during course of normal hexyl resorcinol by mouth

	SUBJECTS																				GRAND TOTALS	PERCENTAGES
	C. B.				C. K.				J. C.				E. C.				V. L.					
	Urine obtained at intervals since last dose																					
	3 hours	5 hours	16 hours	Totals	3 hours	5 hours	16 hours	Totals	3 hours	5 hours	16 hours	Totals	3 hours	5 hours	16 hours	Totals	1 to 3 hours	4 to 5 hours	6 to 8 hours	13 to 20 hours	Totals		
Urine specimens killing both organisms	1	2	1	4	7	4	4	15	7	1	4	12	1	0	3	4	3	0	0	2	5	40	16
Urine specimens killing B. coli only	4	7	3	14	9	8	4	21	6	9	4	19	1	6	3	10	18	10	5	1	34	97	39
Urine specimens killing staphylococcus albus only	1	0	0	1	1	0	1	2	2	1	2	5	0	0	0	0	1	0	1	2	4	13	5
Number of specimens showing bactericidal action	6	9	4	19	17	12	9	38	15	11	10	36	2	6	6	14	22	10	6	5	43	150	60
Number of specimens shown to be inert	11	6	11	28	0	3	6	9	2	4	5	11	15	9	9	33	7	2	2	9	20	101	40
Number of urine specimens examined	17	15	15	47	17	15	15	47	17	15	15	47	17	15	15	47	29	12	8	14	63	251	
Percentage of urine specimens showing bactericidal properties	35	60	27	40	100	80	60	81	88	73	67	76	12	40	40	30	76	83	75	36	68		60

THE SECRETION OF BACTERICIDAL URINE BY NORMAL MEN

Following oral administration of hexyl resorcinol in relatively small doses, normal men secrete urine which, under precisely the same experimental conditions described with rabbit urine, will destroy B. coli and Staphylococcus albus with a high degree of expectancy.

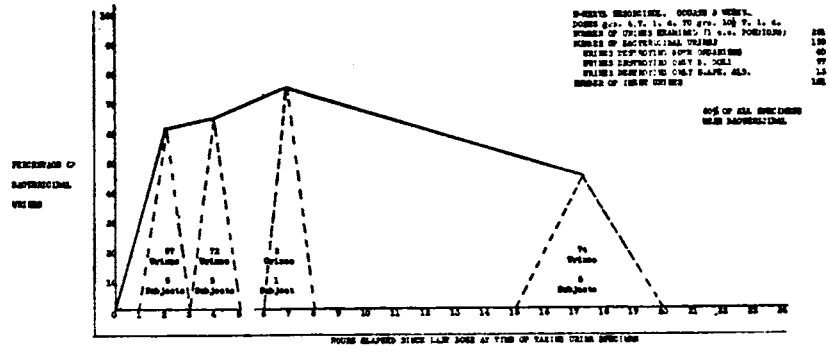


FIG. 3. CURVE BASED ON TABLE 5 SHOWING THE PERCENTAGE OF BACTERICIDAL URINE SPECIMENS OBTAINED AT VARIOUS HOURS FROM FIVE NORMAL MEN DURING A THREE WEEKS COURSE OF N-HEXYL RESORCINOL

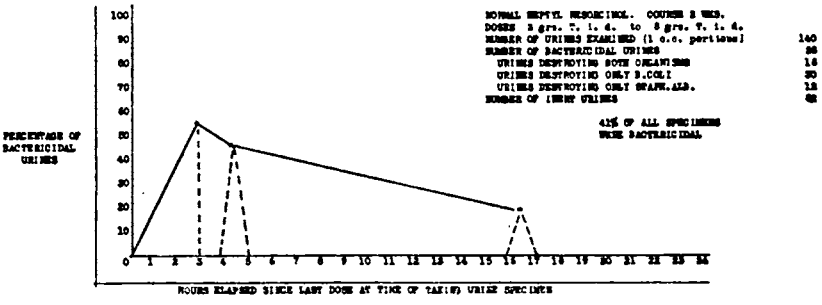


FIG. 4. CURVE BASED ON TABLE 6 SHOWING THE PERCENTAGE OF BACTERICIDAL URINE SPECIMENS OBTAINED AT VARIOUS HOURS FROM FIVE NORMAL MEN DURING A TWO WEEKS COURSE OF N-HEPTYL RESORCINOL

Table 5 summarizes the results obtained during a three weeks course of hexyl resorcinol to 5 normal men in doses of 0.25 to 0.66 gram t.i.d. which were known to cause the secretion of bactericidal urine, while in table 6 the effects of a two weeks course

of heptyl resorcinol in doses of 0.2 to 0.5 gram t.i.d. to the same five men, may be compared. The superiority of hexyl resorcinol in man, as in rabbits, is made very evident in figures 3 and 4 which show the relative expectancy of bactericidal urine in the same 5 normal individuals during courses of each derivative. Sixty per cent of the 251 urine specimens obtained during the course of hexyl resorcinol at intervals of from one to twenty hours after the last dose, were found to destroy one or both test organisms, while this was true of only 41 per cent of the 140 urine specimens obtained at intervals of three to seventeen hours after the last dose of heptyl resorcinol.

Both tables 5 and 6 show very clearly that the expectancy of bactericidal urine under identical experimental conditions may vary widely among normal individuals. The detailed protocols on which these summaries are based show also that the same individual may vary from week to week in this regard. Individual variation is even more striking in rabbits than in man. Two rabbits in my possession have never secreted bactericidal urine in spite of frequent attempts to make them do so by the administration of huge doses of hexyl resorcinol. I have never encountered a normal man who would not secrete bactericidal urine with fair regularity after a day or two on doses of 0.6 gm. hexyl resorcinol three times a day. Most individuals respond promptly in this regard, but one of my subjects (C.B.) requires about three days on this dosage before he begins to secrete bactericidal urine with any regularity. On the other hand I have obtained urine which destroyed both test organisms on doses as low as 0.13 gram in the case of C. K. This subject secretes bactericidal urine with great regularity on minimal doses.

Depending, as it seems we must, upon the elimination of unconjugated hexyl resorcinol in order to obtain bactericidal urine, it is not surprising that some individuals would be found to conjugate this substance more completely than others, or even that the same individual might vary in this regard from week to week. In those individuals like C. B. who conjugate hexyl resorcinol very completely, it is possible nevertheless to accomplish the secretion of free hexyl resorcinol in the urine by the administra-

TABLE 6

Secretion of bactericidal urine by man during course of normal heptyl resorcinol by mouth

	SUBJECTS																				GRAND TOTALS	PERCENTAGES
	C. B.				C. K.				J. C.				E. C.				V. L.					
	Urine obtained at intervals since last dose																					
	3 hours	4 to 5 hours	16 to 17 hours	Totals	3 hours	4 to 5 hours	16 to 17 hours	Totals	3 hours	4 to 5 hours	16 to 17 hours	Totals	3 hours	4 to 5 hours	16 to 17 hours	Totals	3 hours	4 to 5 hours	16 to 17 hours	Totals		
Urine specimens killing both organisms	2	1	0	3	3	1	0	4	2	0	1	3	0	0	2	2	2	2	0	4	16	11.4
Urine specimens killing B. coli only	2	2	1	5	4	4	1	9	2	3	1	6	2	1	1	4	4	2	0	6	30	21.4
Urine specimens killing staphylococcus albus only	0	0	0	0	1	0	1	2	2	1	2	5	1	1	0	2	2	1	0	3	12	8.5
Number of specimens showing bactericidal action	4	3	1	8	8	5	2	15	6	4	4	14	3	2	3	8	8	5	0	13	58	41.0
Number of specimens shown to be inert	7	5	8	20	3	3	7	13	5	4	5	14	8	6	6	20	3	3	9	15	82	59.0
Number of urine specimens examined	11	8	9	28	11	8	9	28	11	8	9	28	11	8	9	28	11	8	9	28	140	
Percentage of urine specimens showing bactericidal properties	36	37	11	28	73	63	22	52	55	50	45	50	27	25	33	28	73	63	0	46		41.0

tion of repeated doses for several consecutive days. It seems likely that under these circumstances the available supply of materials necessary to the conjugation is temporarily depleted, and if the course is continued without interruption the secretion of bactericidal urine continues with a fairly definite expectancy. If, on the other hand, the drug is withheld for twenty-four or forty-eight hours, further treatment for two or three days is again necessary to establish a flow of bactericidal urine.

Two facts brought out by these experiments with hexyl resorcinol in normal men should be emphasized, for the reason that they probably bear some relation to the clinical results to be reported. They apply equally to the hexyl and heptyl derivatives. As shown in table 2, hexyl resorcinol will destroy Staphylococcus albus in far higher dilutions in urine of any reaction than are required to kill B. coli. Yet of the 251 specimens of urine recorded in table 5, 150 of which were bactericidal, 137 killed the colon bacillus while only 53 destroyed the staphylococcus under identical conditions. This difference is far less pronounced in the case of heptyl resorcinol (table 6). On the other hand, many of the 53 specimens of urine bactericidal to Staphylococcus albus completely destroyed this organism after one hour's incubation, while this is a very rare occurrence with B. coli.

Many of these specimens of urine failing to kill one 3 mm. loopful of a twenty-four hour broth culture of Staphylococcus albus, in 1 cc. amounts, completely destroyed this organism if 5 cc. of the same urine were inoculated with approximately the same number of organisms. B. coli, on the other hand, is much less sensitive to the volume of urine employed in the test.

Although detailed information is not yet ready for presentation, it seems highly probable that the velocity of disinfection and the factor of mass action in the disinfection of these organisms by the bactericidal urine concerned, will go far to explain the clinical results obtained with hexyl resorcinol in the treatment of urinary infections, which in some respects were totally unexpected.

Before proceeding to a summary of the clinical results obtained thus far, one other point should be emphasized. Normal

men receiving hexyl resorcinol will not secrete bactericidal urine if sodium bicarbonate is administered with it in sufficient quantity to keep the urine alkaline. This is not due to the alkalinity of the urine *per se*. Specimens of bactericidal acid urine retain their bactericidal properties after titration to alkalinity. In fact they become more active against staphylococci after such titration. Also, alkaline urine secreted spontaneously by either rabbits or man after hexyl resorcinol may be actively bactericidal. It is reasonable to assume as a working hypothesis that sodium bicarbonate in some way facilitates the complete conjugation of hexyl resorcinol in the body. Whatever the explanaton, it is clear that sodium bicarbonate is contraindicated.

RESULTS OF TREATMENT OF URINARY INFECTIONS WITH HEXYL RESORCINOL

Under the conditions obtaining in the experiments described, normal men receiving hexyl resorcinol by mouth secrete urine which will destroy the colon bacillus with a far greater degree of certainty than Staphylococcus albus. On the basis of these results it was predicted that hexyl resorcinol would prove to be more generally efficient in B. coli infections of the urinary tract than in those due to staphylococci. The reverse is true.

On doses of 0.3 to 0.6 gram of hexyl resorcinol three times a day, chronic infections of the urinary tract due to staphylococci and streptococci ordinarily clear up promptly and completely without any other treatment. While the process of disinfection of the urinary tract is in progress in these cases, the symptoms disappear and the urine becomes clear. On complete disinfection of the urinary tract, as shown by a negative culture from 0.5 cc. of bladder urine, it has been my custom to continue treatment with hexyl resorcinol for two weeks. During this period it can be demonstrated that the urine secreted will destroy broth cultures of the strain originally responsible for the infection. No recurrence has been noted as yet in any case treated in this manner, although some have been observed for several months.

When the number of organisms per cubic centimeter of urine is relatively low, chronic pyelitis due to B. coli has been cleared

up completely and permanently with no other treatment than hexyl resorcinol by mouth. A few cases showing rather high counts of B. coli in the kidney urine (400,000 to 500,000 per cubic centimeter) have been cleared up by hexyl resorcinol by mouth combined with renal lavage. On the other hand, those cases of long standing upper urinary tract infections due to B. coli and showing enormous numbers of organisms in the urine with quantities of pus, and perhaps also adiminished phenolsulphonphthalein output, have all been absolute failures in so far as disinfection of the urinary tract is concerned on treatment with hexyl resorcinol alone or combined with the usual procedures.

The fresh bladder urine from a case of chronic pan-urinary B. coli infection not infrequently shows a count of several hundred million organisms per cubic centimeter while in the kidney urine there may be found only a few hundred thousand per cubic centimeter. This of course is due to incubation of the organisms in the bladder. In several cases of this type treatment with hexyl resorcinol alone in some instances, and combined with renal lavage in others, has resulted in complete disinfection of the kidney pelvis, as proved by sterile urine from the renal catheter, while the bladder urine was still teeming with organisms. In one such case still showing many millions of B. coli per cubic centimeter in the bladder urine, after an intensive course of hexyl resorcinol without other treatment, two bladder irrigations with 1–500 silver nitrate resulted unexpectedly in complete disinfection. This patient's urine has since remained sterile without treatment of any kind (four months).

As paradoxical as these clinical results may seem in the light of the experiments on normal men which preceded them, they are nevertheless readily explained. The urine secreted by normal men receiving hexyl resorcinol will destroy the staphylococcus less frequently than the colon bacillus. On the other hand, it will not infrequently destroy the staphylococcus within one hour—a very rare occurrence with B. coli. The velocity of the disinfection of these organisms in the urine may account in part for the clinical results.

It does not require a large clinical experience with hexyl resorcinol, however, to become impressed by the fact that in chronic B. coli infections, the number of organisms present per cubic centimeter of urine is an important guide to the prognosis, in so far as treatment with this drug alone is concerned. Remembering that in urinary infections due to the colon group we are dealing with much larger numbers of organisms per cubic centimeter of urine than in the usual coccus infections and that we must depend for the disfection upon the secretion of small quantities of active hexyl resorcinol in the urine, the factor of masss action in disinfection under these circumstances looms into real significance.

In the coccus infections, the striking clinical results obtained with no other treatment than hexyl resorcinol by mouth prove conclusively that enough of it is eliminated in active form to destroy the relatively small numbers of organisms present. The same is true of B. coli infections with relatively small numbers of organisms present, although persistent treatment is usually necessary even in these cases to complete the disinfection. On the other hand, this amount of free hexyl resorcinol is obviously inadequate to destroy the huge numbers of organisms present in many B. coli infections. Whatever amount is eliminated in the free, unconjugated form is immediately bound and rendered inert in killing off a certain proportion of the bacteria present. The survivors are left in urine which is still a favorable medium and grow actively.

Incubation of a specimen of urine obtained before disinfection is complete will show active growth of the organisms present in spite of the fact that after disinfection of the urinary tract has been accomplished, the urine secreted by the patient still receiving hexyl resorcinol will destroy the organisms originally responsible for the infection.

In practice therefore it has seemed logical, in dealing with chronic B. coli infections of the urinary tract which almost invariably show huge numbers of organisms in the bladder urine, to supplement the administration of hexyl resorcinol with bladder irrigations as a routine. If after a reasonable time the number of organisms in the kidney urine is still high, renal lavage has been

used in an effort to reduce the number present to the point where the hexyl resorcinol might be afforded an opportunity to complete the disinfection. Renal lavage is frequently unnecessary. Bladder irrigations are almost always necessary, unless the B. coli count is very low.

As yet no case of the type described as showing a heavy infection of the kidney urine with quantities of pus and perhaps a subnormal kidney function, has been successfully disinfected with hexyl resorcinol by mouth, alone, or combined with all of the usual procedures. The majority of these cases have shown a pure culture of B. coli in huge numbers. B. pyocyaneus has also been present in a few. It is highly probable that we are dealing in this group with infections which have thoroughly invaded the renal parenchyma. The inference is plain that hexyl resorcinol is inert during its passage through the renal epithelium and this deduction has been supported by direct experimental evidence by an independent observer, which will be reported in due course. All present indications, both clinical and experimental, support the contention that hexyl resorcinol cannot be expected to disinfect the renal parenchyma. It is this observation which originally suggested the idea expressed above, that renal antisepsis and urinary antisepsis must be sharply differentiated, that an internal renal antiseptic should possess experimental qualifications quite dissimilar to those essential to an internal urinary antiseptic, and that it is highly improbable that any one substance could ever satisfy the demands of both fields. Hexyl resorcinol is an internal *urinary* antiseptic.

In view of the fact that the secretion of bactericidal urine follows the administration of hexyl resorcinol by mouth, its use as a prophylactic during investigation of the uninfected urinary tract for diagnostic purposes, is obvious. The experiments on normal men would indicate the desirability of starting its administration a day or two before any instrumentation is undertaken, and continuing it during the period of examination and for a few days thereafter.

The fact that a practically continuous flow of bactericidal urine is readily attainable has suggested another use for hexyl

resorcinol in the prophylaxis and treatment of infections of the operative field following operations, such as prostatectomy, which establish a temporary urinary fistula. If the urinary tract were already heavily infected before operation with an organism such as B. coli for instance, it is safe to assume that the urine would be entirely devoid of any bactericidal properties by the time it reached the operative field. On the other hand, if the urinary tract were uninfected before operation or infected with a Gram-positive organism or B. coli in small numbers, the urine passing over the operative wound would possess active bactericidal properties and might be expected not only to prevent or limit the possibility of wound infection but also to have a beneficial result if the wound infection were already established. Early clinical results in this field have been very encouraging.

In summary, therefore, we have in hexyl resorcinol an internal urinary antiseptic which can be depended upon to clear up infections of the urinary tract by Gram-positive organisms, such as the cocci, without any other treatment. The same holds true for B. coli infections if the bacterial count is low. Local treatment, especially bladder irrigations, is indicated if the B. coli count is high. If the infection has invaded the renal parenchyma extensively (pyelonephritis, etc.) complete disinfection by hexyl resorcinol alone or combined with any known method of treatment is out of the question. There may be pronounced clinical improvement on hexyl resorcinol in heavy B. coli infection without any reduction in the bacterial count in the urine. Detailed clinical reports will be presented in a subsequent communication.

CONCLUSIONS

1. Hexyl resorcinol, a stable organic substance of known chemical constitution, is the most powerful germicide ever described as a non-toxic substance. As measured by the Hygienic Laboratory method now in use it exhibits an increase in bactericidal power over resorcinol, its mother substance, of more than 15,000 per cent, and over phenol of more than 4000 per cent.

2. The application of hexyl resorcinol as an internal urinary antiseptic is based upon a logical and orderly development of the chemical and biological characteristics of its lower homologues.

3. Hexyl resorcinol is non-toxic by mouth and is administrable in repeated doses for indefinite periods. Prolonged administration of large repeated doses to animals and to man results in no injury to the kidney or irritation of the urinary tract. It retains its powerful bactericidal action in solution in human urine of any reaction and is secreted unchanged by the kidney after oral administration in sufficient concentration to impart active bactericidal properties to the urine. No other substance (except its less efficient homologue) has ever been described as possessing these qualifications.

4. The enormous excess of bactericidal power in hexyl resorcinol is necessary to an internal urinary antiseptic of this type because the bulk of each dose is excreted as an inert conjugate. Otherwise a few milligrams a day, instead of 0.3 to 0.6 gram three times a day, would be sufficient to insure a practically concontinuous flow of bactericidal urine.

5. It is highly probable that hexyl resorcinol exerts little if any influence on infections which have invaded the parenchyma of the kidney. It is an internal urinary antiseptic. No internal renal antiseptic is known.

6. Urinary infections due to B. coli and confined to the urinary mucosa from the kidney pelvis downward can be cleared up completely with no other treatment than hexyl resorcinol by mouth if the bacterial count in the urine is low. If the count is high, as is usually the case in B. coli infections, the mass action of the free hexyl resorcinol excreted in the urine is insufficient to complete the disinfection until the count is reduced by local treatment. Persistent treatment (thirty to sixty days) is usually necessary in these cases. If the infection has invaded the renal parenchyma, as is usual in cases of long standing, hexyl resorcinol alone or combined with any known method of treatment will not complete the disinfection and frequently fails even to influence it. In some of these heavy B. coli infections there is, however, very pronounced clinical improvement as evidenced by subsidence of

symptoms, macroscopic clearing of the urine, gain in weight, etc. —without any apparent reduction in the number of organisms per cubic centimeter of urine.

7. Urinary infections due to the usual Gram-positive cocci ordinarily clear up promptly, completely and permanently with no other treatment than hexyl resorcinol by mouth.

REFERENCES

(1) DAVIS, EDWIN: Jour. Amer. Med. Assoc., 1918, lxx, 581.
(2) DAVIS AND WHITE: Jour. Urol., 1918, ii, 107.
(3) DAVIS, WHITE AND ROSEN: Jour. Urol., 1918, ii, 277.
(4) DAVIS, EDWIN: Jour. Urol., 1918, ii, 299.
(5) YOUNG, WHITE AND SWARTZ: Jour. Amer. Med. Assoc., 1919, lxxiii, 1483.
(6) DAVIS, EDWIN: Amer. Jour. Med. Sci., 1921, clxi, 251.
(7) DAVIS, EDWIN: Jour. Urol., 1921, v, 215.
(8) JOHNSON AND HODGE: Jour. Amer. Chem. Soc., 1913, xxxv, 529.
(9) JOHNSON AND LANE: Jour. Amer. Chem. Soc., 1921, xliii, 348.
(10) HAWK, PHILIP B.: Physiological Chemistry. P. Blakiston's Sons & Co., Philadelphia, 1913, 336. Pharm. Zeitg., liv, 612.
(11) DAVIS AND HAIN: Jour. Urol., 1918, ii, 309.