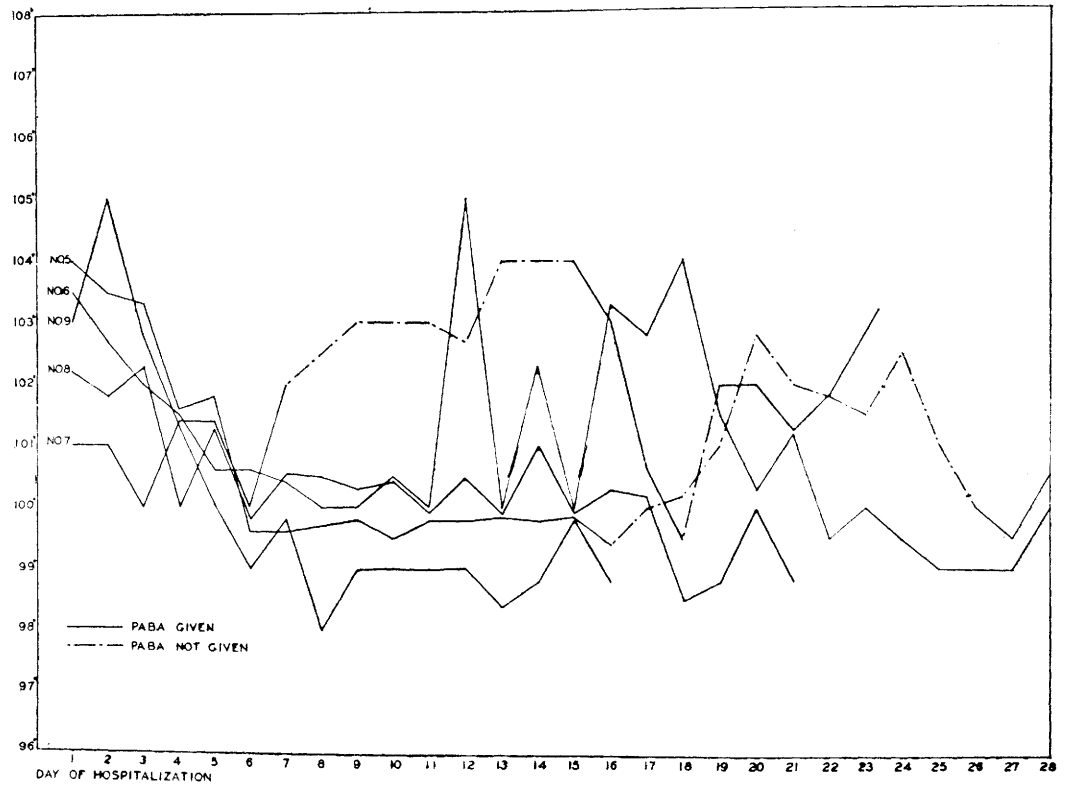


Chart 2.

tion. An unidentified reducing substance was found in the urine in the majority of these cases.

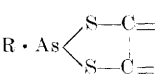
Conclusions. Para-aminobenzoic acid, in the small series of 9 cases of acute rheumatic fever presented, appeared to have a definite effect on the fever and joint pains.

15902

Effect of BAL on Trichomonacidal Activity of Some Organic Arsenicals.

ERNST A. H. FRIEDHEIM AND ROSE L. BERMAN. (Introduced by M. B. Sulzberger.)
From the Laboratory of E. A. H. Friedheim, New York City.

The inclusion of an aromatic trivalent arsenic radical in a 5-membered ring of the type

I 

does not preclude a chemotherapeutic effect with an index of the same order of magnitude, characteristic for the parent compound

II R · AsO

This conclusion was reached in a previous study on the trypanocidal effect of the condensation product of Mapharsen and BAL, *i.e.*, 2-amino-4-[methylolcyclo-(ethylenedimercaptoarsino)]-phenol hydrochloride[1]

1 Friedheim, E. A. H., and Vogel, H. J., PROC. SOC. EXP. BIOL. AND MED., 1947, **64**, 418.

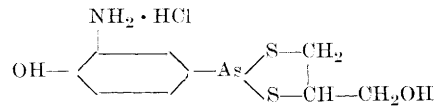
TABLE I.

In Vitro Effect of Some Arsenic and Antimony Compounds and Bromoacetic Acid on *Trichomonas vaginalis.*

Results of Microscopic Dark Field examinations of *Trichomonas vaginalis* from Clinical Cases, Suspended in Drug Solutions.

Drug	Final drug dilution				
	1:100	1:200	1:400	1:600	1:800
Mapharsen BAL[1]	X	X	X	X	0
Mapharsen	0	0	0	0	0
70 A BAL	X	0	0	0	0
70 A	X	0	0	0	0
p-Melaminylphenyl sodium arsonate[2]	0	0	0	0	0
p-Melaminylphenyl sodium stibonate[3]	X	0	0	0	0
Aldarsone	0	0	0	0	0
Acetarsone (Stovarsol)	0	0	0	0	0
Bromoacetic acid	0	0	0	0	0

X = Complete immobilization of trichomonas in less than 5 min.
0 = No or incomplete immobilization in less than 5 min.

III 

in the *T. equiperdum* infection of the mouse.

The same principle has now been found to apply equally well and more so to the immediate toxic *in vitro* effect of III on *Trichomonas vaginalis.*

Observations summarized in Table I show that III not only equals but surpasses significantly the damaging effect of Mapharsen on *Trichomonas vaginalis,* as measured by the drug concentration required to immobilize the parasites within a chosen time interval. Taking 5 minutes as the critical observation time, Mapharsen is inactive in a dilution of 1:100, while its BAL derivative III is active at 1:600.

Considering the possible mechanism of this effect, we note that the condensation with BAL eliminates from the parent molecule a hydrophilic group, *i.e.,* the polar arsenoso radical, and introduces a lipid-soluble group, *i.e.,* the BAL radical. Both changes concur in affecting the water/lipid distribution coefficient in favor of the lipid phase. Thus condensation of Mapharsen with BAL allows for a new distribution of the R · As– radical, permitting the arsenic to come in contact with biochemical systems it could not reach before. The hypothesis may be formed that this new distribution based on relative lipid solubility is responsible for the enhanced toxic effect of the BAL derivative on this particular parasite.

This conception is sustained by the observation on compound 70A, Eagle, (γ-(-arsenosophenyl)-butyric acid), and the corresponding BAL derivative, reported in Table I, although here the condensation with BAL fails to exert a significant effect on the trichomonacidal activity. This result seems at first glance to contradict the hypothesis under discussion, but at closer scrutiny, is well in keeping with it: The butyric acid residue, characteristic for this compound, impresses on the molecule a strongly polar, anionic character, which dominates even after condensation with BAL, precluding the degree of lipid solubility necessary for a trichomonacidal effect.

Table I includes a comparative control of the BAL experiments with 3 standard as well as with 2 new organometallic compounds. It follows that under the chosen experimental conditions, the clinically much used pentavalent arsenic compounds Acetarsone, Aldarsone,* and the metal-free bromoacetic acid

* Acetarsone and Aldarsone failed to immobilize trichomonas even at the highest concentration chosen, *i.e.,* 1:100, within 30 minutes. The mobility of the trichomonas in the suspension continued unabated for 6 hours.

are inactive. Equally inactive is a new arsenical, *p*-melaminylphenyl sodium arsonate,[2] but it is very noteworthy that the analogous antimony compound, *i.e.*, *p*-melaminylphenyl sodium stibonate[3] is of significant activity.

Experimental. Material. Fresh specimens of vaginal secretion from clinical cases of heavy *Trichomonas vaginalis* infestation were suspended in saline. The trichomonas suspension containing 4-12 parasites per field (obj. 20X; ou. 18X) was mixed in test tubes with equal parts of drug solution adjusted to pH 6-7.8. The mixing, the preparation of the slide, and the first microscopic observation consumed on the average 25 seconds. Samples of the mixture were examined after various intervals as to the presence of *mobile* parasites in at least 100 fields.

Criteria for Activity. "Immobility" is here defined as "absence of movement of flagella and undulating membrane" in all parasites encountered. In most preparations, the cessation of movement was followed rapidly by a disintegration of the parasites. In other cases, the immobilization could not with certainty be considered as the expression of a lethal effect. The term "trichomonacidal" is used in the present paper with this restriction.

Compound 70A. γ-(*p*-arsenosophenyl)-butyric acid, was kindly supplied by Dr. Eagle. The BAL derivative was prepared by reacting equal molar quantities of the 70A sodium salt and BAL in an aqueous medium.

The reaction product, *i.e.*, γ-4-[methylolcylco-(ethylenedimercaptoarsino)] - phenylbutyric acid, crystallizes in fine white needles. It is soluble in alkali, ethanol, acetone and propylene glycol, insoluble in water and dilute mineral acids. The nitroprusside test for SH groups is negative at pH 8, but positive in the presence of an excess of free alkali.

Analysis.

Calcd. for $C_{13}H_{17}O_3S_2As$: As, 20.8%
Found : As, 20.5%

Summary. 1. The condensation with BAL enhances significantly the trichomonacidal effect of Mapharsen.

2. The resulting compound, *i.e.*, 2-amino-4-[methylolcyclo-(ethylenedimercaptoarsino)]-phenol, in the form of its hydrochloride, compares favorably with acetarsone, aldarsone, and bromoacetic acid as to trichomonacidal effect.

3. The increase of trichomonacidal activity brought about by condensation with BAL is considered as associated with the concurrent increase in lipophilic character.

4. A comparison between the arsonic and the stibonic acid derived from the identical organic radical shows the arsenic compound to be inactive and the analogous antimony compound to be active and superior to acetarsone and aldarsone as to trichomonacidal effect.

[2] Friedheim, E. A. H., *J. Am. Chem. Soc.*, 1944, **66**, 1775.

[3] Friedheim, E. A. H., Vogel, H. J., and Berman, R. L., *J. Am. Chem. Soc.*, 1947, **69**, 560.