

NOTES

USE OF LIVER EXTRACT AS AN ENRICHMENT FACTOR FOR THE GROWTH OF GONOCOCCI

E. ALTURE-WERBER

Wallace Laboratories, Inc., New Brunswick, N. J.

Received for publication February 5, 1944

It has been shown by Lankford (1942) that certain strains of *Neisseria gonorrhoeae* grow only in the presence of a thermolabile factor which can be derived from blood, fresh liver, and yeast. Lankford and Snell (1943) found that glutamine, an essential growth factor for hemolytic streptococci, helps to detect fastidious strains of gonococci when added to agar base in concentrations of 1-2 mg % (1939) (1940). Peizer and Steffen (1942) have shown that horse plasma

TABLE 1
Proteose agar + 1% glucose (100 ml.)

ADJUVANTS ADDED	STRAINS				
	A	B	C	D	E
0.....	+	+	+	+	+
2 mg Glutamine.....	++	++	++	++	++
5 ml Rabbit cholerae blood.....	++	++	++	++	++
25 ml Liver extract.....	+++	+++	++++	+++	++
10 ml Liver extract.....	+++	+++	++++	+++	++
10 ml Hemolized horse blood.....	+++	++	++++	++++	++
5 ml Hemolized horse blood.....	++	+	++++	++	+++
1 ml Hemolized horse blood.....	++	+	+++	++	+
25 ml Liver extract + 10 cc. horse blood.....	+++	++++	++++	++++	++++
25 ml Liver extract + 5 cc. horse blood.....	++++	++++	++++	++++	+++
10 ml Liver extract + 10 cc. horse blood.....	+++	+++	++++	++++	+++
10 ml Liver extract + 5 cc. horse blood.....	+++	+++	+++	++++	++++

and hemoglobin constitute growth-stimulating enrichments for cultures of gonococci.

It has been found that aqueous solutions of liver extract powder 1/25 (Armour) could be used as an enrichment in gonococcus culture media in place of fresh liver preparations. This product represents the aqueous extract of fresh liver dried *in vacuo* at a low temperature. Processing apparently does not affect the thermolabile factor necessary for the growth of certain strains of *N. gonorrhoeae*. This liver extract alone or in combination with citrated hemolized horse blood proved to be a suitable adjuvant to media used for the cultivation of gonococci.

The various media were prepared as follows:

A 1% suspension of liver extract powder was made in 100 ml of distilled water, thoroughly mixed, filtered through absorbent cotton and then through filter paper. The clear brownish filtrate was sterilized by passage through a Seitz filter and stored in the ice chest until used. Sterilized citrated horse blood was hemolized by freezing. The proteose agar base was autoclaved in 100 ml amounts (4.5 g Difco Bacto Proteose No. 3 Agar in 100 ml of distilled water), after which sterile glucose solution was added to a concentration of 1%. The agar was cooled to 45 C and liver extract and citrated hemolized horse blood were mixed into the agar base and plates were poured. Five strains of *Neisseria gonorrhoeae* were examined. Table 1 shows the results concerning the size of the colonies grown in the liver extract and in hemolized horse blood. The two enrichments were used alone and combined in various amounts with the base media and are compared with colonies grown in the base sugar agar alone, with the addition of 2 mg % of glutamine and with rabbit-blood chocolate agar.

Depending on the amount of hemolized horse blood or liver extract added to the base agar media the cultures grow more or less luxuriantly.

Two mg % glutamine and 5% rabbit chocolate blood give equal results but are less effective than horse blood and liver extract. The best result in the growth for all strains of *Neisseria gonorrhoeae* are obtained when 25 ml of 1% aqueous liver extract and 5 ml citrated hemolized horse blood are added to 100 ml of the proteose sugar agar base.

REFERENCES

- LANKFORD, C. E. 1942 Some aspects of nutritional variation of gonococcus. *J. Bact.*, **44**, 139.
- LANKFORD, C. E., AND SNELL, E. E. 1943 Glutamine as a growth factor for certain strains of *Neisseria gonorrhoeae*. *J. Bact.*, **45**, 410-411.
- McILWAIN, H., FILDES, P., GLADSTONE, G. P., AND KNIGHT, B. C. J. G. 1939 Glutamine and the growth of *Streptococcus hemolyticus*. *Biochem. J.*, **33**, 223-229.
- FILDES, P., AND GLADSTONE, G. P. 1940 Glutamine and the growth of bacteria. *Brit. J. Exptl. Path.*, **20**, 334-341.
- PEIZER, L. R., AND STEFFEN, G. I. 1942 A modification of the horse plasma hemoglobin agar for primary culture of the gonococcus. *Venereal Disease Information*, **23**, 224-226.

SUBSTITUTES IN CULTURE MEDIA

V. BENNETT McMAHON

University of California Medical Center, San Francisco

Received for publication February 21, 1944

Three stable, economical culture media which can be duplicated successfully and which contain substitutes for products difficult to get are described. Two contain substitutes for meat; the third, a substitute for tomatoes. The media have been tested in teaching, clinical, and research laboratories.

A medium, superior to meat in nutritive properties, has been made from soy beans. The formula adopted follows: