Prothrombin Activity of Blood of the Newborn.*† (20143)

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It has been repeatedly observed that the blood of the newborn baby has a normal prothrombin time, provided the mother is supplied with vit. K before the baby is born, but a prothrombin concentration of only about 30% when measured by the two-stage method. Brinkhous, Smith and Warner(1) originally explained this discrepancy by postulating a compensatory acceleration of the conversion of prothrombin in newborn blood. More recently this acceleration is being attributed to specific conversion factors(2). If this explanation is valid, the plasma of a newborn baby should contain a relatively high concentration of the accelerator and a low amount of prothrombin, whereas normal adult blood should have 3 times more prothrombin and a corresponding lower concentration of the accelerator. If the prothrombin time is dependent not only upon prothrombin, but also on the accelerator factor, one should expect that when adult and newborn baby plasmas are mixed in varying proportions a detectable change in prothrombin time should occur since the ratio of prothrombin to accelerator can be greatly varied. From the results recorded in Table I it is clear that in a series of mixtures ranging from a ratio of 9 parts of newborn to 1 part of adult plasma to a reverse ratio of 1 to 9, no demonstrable change in the prothrombin time was observed.

To obtain further information concerning the relative prothrombin activity of newborn plasma, the effect of storage in glass was investigated. When normal adult plasma is stored at 4°C in glass, the prothrombin time which initially is 12 seconds becomes prolonged due to loss of labile factor. On restoring the latter by adding deprothrombinized rabbit plasma which serves as a rich source, the prothrombin time becomes shortened to about 8 to 9 seconds. The prothrombin time of newborn plasma likewise becomes prolonged on storage, but restoration of the labile factor brings the prothrombin time only to 12 seconds. When stored plasmas of the newborn and of the adult were mixed in varying proportions and deprothrombinized rabbit plasma added, the prothrombin times of the mixtures corresponded with the values expected on the basis of the prothrombin curve(3).

It is experimentally established that a decrease of either prothrombin or labile factor prolongs the prothrombin time. Since the concentration of labile factor is approximately the same in newborn as in adult plasma(4), it is unlikely that the agent is the accelerator hypothesized to explain the observed normal prothrombin time. The present results furnish no support for an accelerator, and a search of the literature fails likewise to disclose any convincing evidence that such a compensating converting factor exists in newborn blood. Such an agent was postulated to account for the seemingly paradoxical situation of a normal prothrombin time with a prothrombin as measured by the 2-stage procedure of less than one-third of normal. By correlating the present results with the previous findings(4) that newborn blood has the same concentration of active prothrombin as adult blood, but contains little or no inactive prothrombin (prothrombinogen), which in adult blood constitutes the largest fraction of the total prothrombin, the discrepancy in findings between the one and two stage methods is readily explained without introducing hypothetical accelerator or conversion factors.

Summary. When adult and newborn plasmas were mixed in varying proportions, the prothrombin time which was 12 seconds for each plasma remained unaltered in the series of mixtures. This suggests that the prothrombin activity *i.e.*, the concentration of active prothrombin, is the same in the plasma of the

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TABLE I. The Effect of Mixing Plasmas of Newborn and Adult on Prothrombin Time.

Fresh cord plasma	cc	.1	.09	.08	.07	.06	.05	.04	.03	.02	.01	.0	
" adult "	cc	—	.01	.02	.03	.04	.05	.06	.07	.08	.09	.1	
Prothrombin time*	sec.	12	12	12	12	12	12	12	12	12	12	12	
Stored cord plasma	cc	.1	.1	.09	.08	.07	.06	.05	.04	.03	.02	.01	— —
" adult "	cc	—	—	.01	.02	.03	.04	.05	.06	.07	.08	.09	.1 .1
Deprothrombinized rabbit plasma	cc	—	.02	.02	.02	.02	.02	.02	.02	.02	.02	.02	.02 —
Prothrombin time*	sec.	24	12	10½	10	9½	9	8½	8½	8¼	8	8	8 35

* The method employed has been previously described(3).
The mothers were supplied with vit. K (synkayvite) prepartally.

newborn baby as in that of the adult.

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Growth of Poliomyelitis Viruses in Large Stationary Cultures.* (20144)

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The first reported methods for propagating poliomyelitis viruses in tissue cultures utilized implants of the suspended cell type(1). Subsequently roller tubes in which the proliferating fragments of tissue were imbedded in plasma clots were used(2). In addition to permitting direct microscopic visualization of cultures for cytopathogenic effect, this method had the advantage of giving an early growth of virus(3). Recent reports have indicated that these same advantages obtain if culture tubes are kept stationary instead of rotated(4,5).

In this report are presented results obtained with the growth of poliomyelitis viruses in large stationary flask cultures. A simplified medium (Hanks-Simms) and fragments of monkey testes imbedded in plasma clots were used. The work was done in order to obtain large quantities of supernatant fluid of sufficiently high virus titer for complement-fixing antigen preparation according to the method of Svedmyr and Enders(6) without having to maintain many small culture units and without having to use special equipment for rotating large culture bottles.

Materials and methods. Viruses. Brunhilde (type 1), Y-SK (type 2) and Saukett† (type 3) strains of poliomyelitis viruses were used in titered pools of supernatant fluid from infected roller tube cultures of monkey testes. Each virus strain had been through at least three consecutive tissue culture passages, and was specifically neutralized by prototype antiserums. *Chicken plasma.* Dehydrated chicken plasma (Difco) was reconstituted and carbonated in the manner reported by Melnick (7). *Monkey testes.* Testes from 3 to 5 normal immature *rhesus* (*M. mulatta*) monkeys were minced together and used for cultures on the same day they were obtained. *Nutrient fluid.* Two kinds of fluid were used. One referred to as "complete medium" contained Hanks and Simms solutions and acetone-extracted chick embryo extract(7). The

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