the marked deposition of numerous masses of non-cellular deeply staining pigment granules (Fig. 4). These granules were even more prominent in the spleens of fish infected with fin-rot disease (Fig. 5).

Discussion. In these experiments the target organ did respond differentially to CA and DCA. In agreement with the results of Bern(1) for *T. mossambica,* CA caused hypertrophy of the spleen and the proliferation of reticular elements. These results would not be expected on the basis of mammalian studies(5).

L. machrochirus differs from *T. mossambica* in that administration of DCA caused atrophy of the spleen and a marked deposition of pigment granules. Thus, a differential response is seen, but the meaning of the response is difficult to evaluate. The nature of the corticoids produced by *L. machrochirus* is not known; but it is probable on the basis of studies carried out on other teleosts(6) that DCA would not be a natural steroid for this animal. It also appears that the dosage given was close to a minimum one required to evoke a response, for a pilot experiment in which DCA was given on alternate days for a 10-day period failed to evoke a response. Further, neither prolonged cold stress nor fin-rot disease duplicated the type of response obtained by administration of cortical steroids.

The blue-gill also differs from *T. mossambica* in that a consistent response to CA was a marked dilation of splenic vessels. This response, along with a proliferation of reticular elements, may account for the enlargement of the spleen. Dilation of splenic vessels is considered to be a direct basis for hypertrophy of mammalian spleens(7). As shown by the Group I animals, phagocytic proliferation may occur during the course of splenic atrophy.

Summary. Administration of CA for 10 or 14 days caused splenic hypertrophy, proliferation of phagocytic elements, and dilation of splenic vessels. Treatment with DCA for the same period caused splenic atrophy and deposition of pigment granules. These granules were also prominent in spleens of animals infected by a fungus disease. Holding the animals at 4°C for 14 days caused splenic atrophy and the appearance of phagocytic elements. A 4-minute exposure to 4°C for 7 days had no effect on spleen weight or histology.

1. Bern, H. A., Proc. Soc. Exp. Biol. and Med., 1963, v112, 805.
2. Krauter, D., Arch. Entw.-Mech., 1958, v150, 607.
3. Holmes, W. N., Acta endocrin., 1959, v31, 587.
4. Yoffey, J. M., J. Anat., 1929, v63, 314.
5. Selye, H., Stress, Acta Publishing Co., Montreal, 1950, p476.
6. Chester Jones, I., Phillips, T. G., Holmes, W. N., Comparative Endocrinology, A. Gorbman, Ed., John Wiley & Sons, Inc. N. Y., p582.
7. Ferry, C. B., J. Physiol., 1962, v162, 65.

Received June 25, 1965. P.S.E.B.M., 1965, v120.

Effects of Autologous and Homologous Blood Replacement in Hypovolemic Dogs.* (30485)

E. L. SMITH, S. DEAVERS AND R. A. HUGGINS
Departments of Physiology, Baylor University College of Medicine and University of Texas Dental Branch, Houston

A number of different effects have been reported in the dog if homologous, rather than autologous, blood or plasma transfusions were given, but the only effect that has been substantiated in our laboratory was the development of urticaria(1-4). The incidence of urticaria in dogs receiving a homologous overtransfusion was 10% with fresh whole blood, 70-90% with fresh plasma or with previously

* This study was supported, in part, by a grant from Nat. Heart Inst., USPHS HE 08793 and, in part, by the Medical Research and Development Board, Dept. of the Army, Contract DA-49-193-MD-2357.

frozen plasma, and 100% with whole blood if it was separated into cell and plasma fractions by the usual centrifugation procedure, then reconstituted and transfused. In all instances the transfusions were in addition to the dog's original blood volume(4). While no definitive study was done at the time, a few experiments were performed whose results suggested that if the blood or plasma given represented a replacement rather than an overtransfusion the incidence of urticaria might be less. The present experiment was designed to investigate this possibility and to compare the effectiveness of homologous and autologous blood in restoring the blood volume in hypovolemia.

Methods. Experiments were conducted on 32 morphine-pentobarbitalized dogs ranging in weight from 10.5 to 18 kg. Plasma volume was determined by the I^{131}-tagged albumin method. Samples were drawn from the right jugular vein through an indwelling cardiac catheter immediately before and 10, 20 and 30 minutes after injection of the isotope. The general procedures are described in detail elsewhere(5). Plasma protein concentration was calculated from the specific gravity determined by the falling drop method(6). Hematocrits (Wintrobe) were determined on all blood samples. Mean carotid arterial pressure was followed throughout the experiments by a mercury manometer.

The dogs were divided into 4 groups of 8 dogs each and each served as his own control. After control measurements the animals were bled from a femoral artery until their mean arterial blood pressure reached 50 mm Hg. Then the bleeding was stopped and one group was immediately reinfused with the shed blood through a femoral vein (autologous). Another group was reinfused with their own blood which had been centrifuged, separated into cells and plasma, and reconstituted (reconstituted autologous). A third group received an infusion of blood from a donor dog (homologous) after their blood pressure reached the 50 mm Hg level, and finally a fourth group received reconstituted homologous blood as a replacement. The time intervals from the start of bleeding to the end of reinfusion were 37 minutes for the autologous, 58 minutes for the reconstituted autologous, 46 minutes for the homologous, and 55 minutes for the reconstituted homologous group. Twenty minutes were allowed for the infused blood to equilibrate and then final determinations were made. The last blood sample was drawn 60 minutes after the second isotope injection, or 80 minutes after the infusion was completed.

Results and discussion. Autologous: Table IA. Twenty minutes after the reinfusion of the blood that was removed (39.7 ml/kg) to lower this group's mean arterial pressure to a level of 50 mm Hg, there was a significant increase (11%) in the venous hematocrit. Plasma volume and both the concentration and total amount of protein were restored to the initial levels. The mean arterial pressure increased by 4% after restoration of the blood volume and was maintained at this level during the following 60 minutes. Reconstituted autologous blood replacement had similar effects on the venous hematocrit which was 11% higher than the expected (Table IB). There was a slight reduction in plasma volume of 2.9 ml/kg and a 7% decrease in total protein although the protein concentration did not change. Twenty minutes after the replacement of blood, the mean arterial pressure was at the initial level, but subsequently there was a 6% reduction in pressure during the next hour. Urticaria was not seen during or after autologous and reconstituted autologous blood infusion.

Homologous blood replacement (Table 2A) resulted in an 18% increase of the venous hematocrit above the "expected." Plasma volume and concentration and total amount of protein were reduced, but not significantly, by 4 and 5%, respectively. Twenty minutes after homologous blood replacement, the mean arterial pressure was 8% less than the initial pressure, it subsequently fell by 13%, and at the end of the experiment it was 12% less than the initial pressure. With homologous blood, 25% of the dogs developed urticaria during or after the blood replacement. In spite of the urticaria, homologous, like autologous, blood effectively restored the blood volume. A similar finding has been reported with homologous plasma(7).

After a hemorrhage to a mean arterial pressure of 50 mm Hg, transfusion of reconstituted homologous blood resulted in a reduction in mean arterial pressure from the initial

TABLE I. Hemorrhage to 50 mm Hg Mean Arterial Pressure and Immediate Replacement with Autologous Blood.

	Venous hematocrit, %	Plasma vol, ml/kg	Plasma proteins g/100 ml	Plasma proteins g/kg	Mean arterial pressure, mm Hg
		A. Autologous blood*			
Control	46.9 ± 1.08	44.4 ± 1.31	5.56 ± .32	2.47 ± .17	100 ± 5.0
Hemorrhage to 50 mm Hg (39.7 ± 3.2 ml/kg)					
Replacement blood	48.2 ± 1.88	21.2 ± 1.57	5.35 ± .30	1.15 ± .12	50
After replacement:					
Expected	46.9 ± 1.08	44.4 ± 1.31	5.56 ± .32	2.47 ± .17	
Measured	52.6 ± 1.84	44.3 ± 1.94	5.48 ± .30	2.44 ± .19	104 ± 8.1
50 min later	52.4 ± 2.13		5.49 ± .32		104 ± 8.1
80 " "	53.2 ± 2.21		5.49 ± .30		103 ±10.6
		B. Reconstituted autologous blood†			
Control	41.2 ± 1.50	47.8 ± 1.54	5.97 ± .17	2.85 ± .13	98 ± 3.7
Hemorrhage to 50 mm Hg (37.1 ± 1.7 ml/kg)	39.6 ± 1.50	22.4 ± 1.10	5.89 ± .14	1.32 ± .08	50
Replacement blood	38.1 ± 1.47	20.0 ± 1.00	5.84 ± .14	1.17 ± .08	64 ± 6.3
After replacement:					
Expected	40.6 ± 1.50	45.4 ± 1.59	5.95 ± .16	2.70 ± .13	
Measured	45.5 ± 1.83	42.5 ± 2.10	5.92 ± .19	2.52 ± .17	97 ± 7.5
50 min later	45.7 ± 1.96		5.79 ± .18		95 ± 9.8
80 " "	45.6 ± 2.07		5.77 ± .18		92 ±11.1

* Values are means with S.E. for 8 dogs with a mean weight of 12.9 ± .56 kg.
† Values are means with S.E. for 8 dogs with a mean weight of 12.9 ± .87 kg.

TABLE II. Hemorrhage to 50 mm Hg Mean Arterial Pressure and Immediate Replacement with Homologous Blood.

	Venous hematocrit, %	Plasma vol, ml/kg	Plasma proteins g/100 ml	Plasma proteins g/kg	Mean arterial pressure, mm Hg
		A. Homologous blood*			
Control	40.3 ± 1.18	47.1 ± 1.96	4.40 ± .20	2.07 ± .11	101 ± 3.7
Hemorrhage to 50 mm Hg (42.2 ± 2.4 ml/kg)	41.1 ± 1.15	25.7 ± 1.34	4.25 ± .19	1.09 ± .04	50
Replacement blood	43.8 ± 1.44	23.7 ± 1.34	4.78 ± .15	1.14 ± .09	
After replacement:					
Expected	42.1 ± 1.50	45.1 ± 2.05	4.69 ± .12	2.12 ± .13	
Measured	51.3 ± 2.67	43.3 ± 3.83	4.50 ± .15	2.01 ± .20	93 ± 4.8
50 min later	51.9 ± 2.78		4.39 ± .13		88 ± 5.2
80 " "	51.2 ± 3.08		4.45 ± .16		89 ± 5.7
		B. Reconstituted homologous blood†			
Control	41.1 ± 2.44	45.8 ± 3.01	4.73 ± .14	2.17 ± .16	103 ± 2.7
Hemorrhage to 50 mm Hg (35.6 ± 3.4 ml/kg)	43.8 ± 2.53	21.5 ± 2.58	4.65 ± .12	.96 ± .10	50
Replacement blood	40.9 ± 1.83	21.2 ± 2.62	4.17 ± .16	.90 ± .13	
After replacement:					
Expected	39.5 ± 1.85	45.5 ± 2.63	4.61 ± .13	2.11 ± .16	
Measured	52.9 ± 2.17	37.9 ± 2.63	4.74 ± .11	1.80 ± .14	96 ± 4.5
50 min later	53.6 ± 2.47		4.68 ± .09		90 ± 8.2
80 " "	53.3 ± 2.35		4.77 ± .06		88 ± 8.2

* Values are means with S.E. for 8 dogs with a mean weight of 11.6 ± .33 kg.
† Values are means with S.E. for 8 dogs with a mean weight of 14.0 ± .48 kg.

level; 7% at 20 minutes, 13% at 50 minutes, and 15% at 80 minutes after the replacement (Table 2B). There was a significant loss of 7.6 ml/kg of plasma ($p<0.005$) and 0.31 g/kg of protein ($p<0.01$) from the circulation. Protein concentration increased slightly, but not significantly. The greatest change was in the venous hematocrit which increased by 25% above that expected ($p<0.001$). Part of the increase was due to the loss of plasma; however, when an "expected" venous hematocrit was calculated based on this loss, the measured venous hematocrit was still 15% higher than the "expected." This could have resulted from the addition of a significant amount of red cells from a previously unmeasured pool or from a shift of either red cells or plasma from one blood compartment to another within the circulatory system. Cell volume determinations were not made in these dogs since there is adequate evidence that the measured and "expected" red cell volumes are in good agreement after a reinfusion following a hemorrhage to 50 mm Hg arterial pressure(8), after an exchange transfusion of either autologous or homologous whole blood(9), or homologous plasma(10), or after an overtransfusion of less than 100 ml/kg of homologous whole blood(11). Therefore, a change in the distribution of cells and plasma between the large and small vessels must have occurred to produce the significantly elevated venous hematocrit in these dogs.

Previous experiments clearly indicate that handling of blood results in an increased incidence of urticaria when transfused into a dog other than the one from which it was obtained. The role of overtransfusion in its production is not clear. In these studies, when reconstituted homologous blood was used, 100% of the dogs developed urticaria (4). In the current experiment 25% of the dogs receiving homologous blood and 63% receiving reconstituted homologous blood developed urticaria. The difference between 100% and 63% in the incidence of urticaria with reconstituted homologous blood suggests that an overtransfusion may facilitate the development of urticaria. These experiments further support previous data that handling of blood (centrifugation, separation and reconstitution) produces a substance causing urticaria. Further, reconstituted blood, whether autologous or homologous, caused the loss of fluid and protein from the circulation. This was particularly evident after reconstituted homologous blood replacement.

Summary. Replacement of blood after a hemorrhage to 50 mm Hg mean arterial pressure with autologous, reconstituted autologous, and homologous blood results in a significantly higher venous hematocrit than expected, indicating shifting of red cells and plasma within the circulation. Mean arterial pressure, plasma volume, and concentration and total amount of protein were not significantly different from the "expected" values. When reconstituted homologous blood was used as the replacement there was a significant reduction in plasma volume and total protein in addition to a steep rise in venous hematocrit. Urticaria was seen in 25% of the dogs receiving homologous blood and in 63% receiving reconstituted homologous blood, supporting the idea that handling of the blood to be transfused produces a substance causing urticaria. In the present experiments reconstituted blood, whether homologous or autologous, caused the loss of fluid and protein from the circulation.

1. Bliss, J. Q., Johns, D. G., Burgen, A. S. V., Circulation Res., 1959, v7, 79.

2. Remington, J. W., Baker, C. H., Am. J. Physiol., 1959, v197, 193.

3. Hutchison, J. L., Freedman, S. O., Richards, B. A., Burgen, A. S. V., J. Lab. Clin. Med., 1960, v56, 734.

4. Huggins, R. A., Smith, E. L., Deavers, S., Am. J. Physiol., 1965, v209, in press.

5. ———, ibid., 1963, v205, 351.

6. Barbour, H. G., Hamilton, W. F., J. Biol. Chem., 1926, v69, 625.

7. Elias, G. L., Gurd, F. N., Hampson, L. G., Burgen, A. S. V., Circulation Res., 1962, v11, 857.

8. Deavers, S., Smith, E. L., Huggins, R. A., Am. J. Physiol., 1958, v195, 73.

9. Deavers, S., Huggins, R. A. Smith, E. L., ibid., 1965, in press.

10. Huggins, R. A., Deavers, S., Smith, E. L., Proc. Soc. Exp. Biol. and Med., 1964, v115, 36.

11. Smith, E. L., Deavers, S., Huggins, R. A., Am. J. Physiol., 1961, v201, 383.

Received June 28, 1965. P.S.E.B.M., 1965, v120.