GLOBIN UTILIZATION BY THE ANEMIC DOG TO FORM NEW HEMOGLOBIN

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In all work relating to hemoglobin one unconsciously has in mind the pigment radicle as being of prime importance because it is a conspicuous factor due to its color and its relation to other body pigments (bile pigment, urobilin and related substances). The pyrrol aggregate—a compound of four pyrrol rings—is common to bile pigment and to the hemoglobin pigment radicle.

Globin by contrast has been little studied but its importance probably is in proportion to its size and it makes up about 95 per cent of the hemoglobin molecule. It is antigenic (4) and its chemical make-up has been studied but we know very little about its manufacture within the body and what happens when it is broken down in body metabolism.

One can readily show (6) that the anemic dog can use goose and sheep hemoglobin just as completely as dog hemoglobin when given intravenously, and if we give 50 gm. of either foreign hemoglobin we confidently expect to remove from the standardized anemic dog 45 to 50 gm. new dog hemoglobin to maintain the anemia level a constant. In a great number of experiments the return averages approximately 100 per cent—that is the dog conserves completely hemoglobin given by vein whether the hemoglobin derives from the dog, goose or sheep. As hemoglobin is species specific it is obvious that the foreign hemoglobin must be broken down to some types of intermediates before it is recast into the new dog hemoglobin.

Other experiments (3) indicate the importance of the *globin fraction* in contrast to the pigment radicle. If to an anemic *bile fistula* dog we give by vein 50 gm. of dog or sheep hemoglobin, we observe a return in new hemoglobin of 45 to 50 gm. just as in the anemic non-

fistula dog. But we observe also a great increase in bile pigment eliminated and can account for 60 to 100 per cent of the pigment radicle contained in the injected hemoglobin. Whatever our explanation may be the body obviously can produce a large excess of the pigment radicle promptly whether it is eliminated in the urine as bile pigment or incorporated in the new formed hemoglobin. This experiment would point to the *globin* as the limiting factor in new hemoglobin production and would indicate the need for a detailed study of all aspects of globin utilization within the body.

We should keep in mind the fact that the anemic fasting dog when given iron by mouth or vein can produce 100 gm. or more of new hemoglobin. The globin and pigment radicles obviously are formed from the proteins within the body (tissue, parenchyma or plasma proteins) (2). This effort by the fasting anemic dog cannot be long continued but the proof is conclusive that in this emergency (anemia) the body proteins can contribute to the formation of new hemoglobin and its contained globin.

Our interest in the reaction of the anemic dog to *globin* given by mouth or by vein needs no further comment. The tables below indicate that the standard anemic dog in order to form hemoglobin utilizes *globin* by mouth to good advantage. Globin given orally averages 30 per cent utilization in contrast to a 10 to 15 per cent utilization of hemoglobin given by mouth.

Globin given by vein does not give a uniform reaction as is true for hemoglobin given by vein. The reasons for this difference in reaction may be subject to debate. Hemoglobin is a naturally occurring substance in the circulation as red cells disintegrate from normal wear and tear. Globin as we use it to introduce into the circulation is probably a foreign substance as it is often toxic in small doses and always toxic in large doses. In other words the breakdown of hemoglobin within the body produces a slightly different substance than the globin produced by acid acetone precipitation in the method used below.

Globin prepared from dog or horse hemoglobin given by vein can be well utilized in certain experiments to form new hemoglobin in the standard anemia dog (Tables 1 to 3). The surplus hemoglobin appears usually in the period following the injection of globin and

there may actually be some inhibition of hemoglobin production during the period of injection. The intoxication (severe, mild or non-recognizable) associated with globin injection is probably responsible for this delay and may explain the negative experiments. We have shown elsewhere (5) that the intoxication associated with a sterile abscess or endometritis will temporarily interfere with the internal metabolism responsible for new hemoglobin production in anemia.

Methods

All method details relating to the general anemia program have been described quite recently (7) and need not be repeated. Globin was prepared from horse and dog hemoglobin according to the method of Anson and Mirsky (1) which is essentially a precipitation by a solution of acetone and hydrochloric acid.

We are indebted to Dr. A. E. Mirsky for a careful explanation of this method and a generous supply of the globin which was used in preliminary experiments. The globin as used is a grey-white powder readily soluble in water with dextrose for intravenous use. It is probably digested with relative ease when fed with the standard salmon bread.

The artificial *digest* used in Table 1 and given by vein was prepared as follows: Horse globin 60 gm. in 3000 cc. 0.2 per cent HCl placed in incubator for 4 days. Solution was neutralized to phenolphthalein with NaOH and 2 gm. each of trypsin and erepsin was added. Material kept in incubator for 4 weeks. Reaction was kept alkaline to phenolphthalein. Hydrolysate was filtered and concentrated to 800 cc. under diminished pressure at about 40°C. Concentrated NaOH was added and NH_3 was removed by distillation under diminished pressure at temperature of 40°. Hydrolysate was immediately treated with 10 per cent HCl until reaction was just alkaline to litmus. Solution was sterilized in autoclave for 30 min. at 15 pounds pressure and kept at ice box temperature. No precipitation on standing. This hydrolysate was prepared under the supervision of Dr. F. S. Daft.

EXPERIMENTAL OBSERVATIONS

Unfortunately solutions of globin given by vein are somewhat toxic and at times may be very toxic. Small doses of globin must be given (1 gm. usually dissolved in 10 per cent dextrose) and in spite of every precaution and slow injection into the jugular we may observe at times clinical disturbances (fall in blood pressure, chill, vomiting or prostration) and on rare occasions moderately severe shock. Dog globin appears to be a little less toxic than horse globin. As the

standardized dogs are valuable we hesitated to use larger doses for fear of severe or lethal intoxication.

Table 1 (dog 27-236) shows three satisfactory experiments on the same dog at different times. In the first experiment *horse globin* is given *intravenously* over a period of 3 weeks amounting to 21 gm. in all. The dog was not clinically disturbed and ate all of the basal ration. There was a rise in hemoglobin level during the period of injection but no bleeding was done at this time. The new hemoglobin production was completed by the end of the 2nd control week. We note a net hemoglobin production above the basal output of 24 gm. new hemoglobin as a result of injecting 21 gm. globin.

Globin digest (Table 1) shows a similar reaction. When we give intravenously a tryptic digest derived from 18.3 gm. horse globin during a 2 weeks period we note a production of 25 gm. new hemoglobin above the base line output. There is a slight decrease in basal ration consumption and one might expect a lessened output of hemoglobin because of this slight intoxication (5). The fact that there is a slight excess of hemoglobin production above the globin digest intake we are not inclined to stress as it comes within the limits of physiological variables.

In general the tables below show good evidence that the anemic dog can use dog or horse globin given by vein to produce much new hemoglobin. The reaction often may be delayed and occasionally absent but we believe this is to be explained in large part as due to the toxic reaction of the dog to the artificially prepared globin solution.

Clinical Experimental Histories—Table 1.

Dog 27-236. Born Feb., 1928. Uneventful anemia history Apr., 1930, to Aug., 1935.

June 10, 1931. Horse globin 0.66 gm. to 1 gm. injected intravenously daily in 75 to 150 cc. 10 per cent dextrose solution. Material injected very slowly. After 11 doses dog showed moderate diarrhea and slight vomiting for 1 day. No further reaction.

Apr. 12, 1932. Globin digest by vein beginning with 10 cc. of the digest or the equivalent of 0.5 gm. horse globin plus 40 cc. 10 per cent dextrose. 3 hours after injection of first dose dog vomited. Dose was increased thereafter from 0.75 gm. equivalent to 2 gm. equivalent daily with from 40 to 100 cc. 10 per cent dextrose solution. After first 2 gm. equivalent dose dog left 70 per cent food for 1 day.

TABLE 1

Globin (Horse) Given by Vein and by Mouth

A Globin Digest also Utilized by Dog to Form Hemoglobin

Dog 27-236. Bull, male, adult.

Diet periods 1 wk. each	Food consumed	Weight	Plasma volume	R.B.C.	Blood Hb. level	Hb. removed, bled
Food, gm. per day	*per cent*	*kg.*	*cc.*	*mil.*	*per cent*	*gm.*
Bread 375, salm. 100, Kl. 30	100	13.4	894	4.6	48	1.3
*Globin 1 intrav., br. 375, salm. 100, Kl. 30	100	13.2	822	6.1	68	2.9
Globin 1 intrav., br. 375, salm. 100, Kl. 30	100	13.4	1028	6.9	67	2.9
Globin 1 intrav., br. 375, salm. 100, Kl. 30	100	13.4	825	5.7	61	3.6
Bread 375, salm. 100, Kl. 30	100	13.3	794	5.3	63	28.2
Bread 375, salm. 100, Kl. 30	100	13.2	734	5.3	41	40.6
Bread 375, salm. 100, Kl. 30	100	13.1	782	4.0	41	1.2
Basal output 8 gm. Hb. per wk. Total net Hb. output 24 gm. Total Hb. = 79.4						
Bread 400, salm. 100, Kl. 40	100	16.1	934	5.4	48	1.3
†Globin digest intrav., br. 400, salm. 100, Kl. 40	91	15.6	855	6.4	55	1.6
Globin digest intrav., br. 400, salm. 100, Kl. 40	91	16.0	863	6.7	59	14.6
Bread 350, salm. 150, Kl. 50	90	15.9	848	6.8	51	26.2
Bread 400, salm. 150, Kl. 50	100	16.0	889	5.7	46	13.7
Bread 400, salm. 100, Kl. 40	100	16.1	894	5.6	45	12.2
Basal output 8 gm. Hb. per wk. Total net Hb. output 25 gm. Total Hb. = 68.3						
Bread 375, salm. 125, Kl. 50	100	16.0	938	4.7	44	1.2
Globin 6, br. 375, salm. 125, Kl. 50	100	16.3	893	5.2	51	16.5
Globin 6, br. 375, salm. 125, Kl. 50	100	16.6	936	5.9	38	34.7
Bread 375, salm. 100, Kl. 40	100	16.4	976	4.3	38	1.8
Bread 400, salm. 100, Kl. 40	100	16.8	1000	5.2	44	1.2
Basal output 5 gm. Hb. per wk. Total net Hb. output 34 gm. Total Hb. = 54.2						

Salm. = commercial canned salmon.

Kl. = Klim, a commercial skimmed milk powder.

* Total globin given 21 gm.

† Total globin equivalent 18.3 gm.

No further unfavorable reaction. Total amount given equivalent to 18.3 gm. horse globin.

Sept. 30, 1933. Globin feeding begun with daily dose of 6 gm. mixed with small amount of bread and fed prior to general feeding.

Globin feeding (Table 1) in the same dog gives about a 40 per cent return of new hemoglobin. The dog was fed 6 gm. horse globin a day for 2 weeks—a total of 84 gm. and produced a net hemoglobin output of 34 gm. above the base line average. This dog gave a very prompt response to the globin feeding and most of the new formed hemoglobin appeared during the 2 weeks of globin feeding. This represents a maximal response to protein material given by mouth and exceeds considerably the ratio of *liver protein* intake to new hemoglobin output—approximately 8 to 1 or a 13 per cent utilization, if we leave out of consideration the iron contained in the liver.

Clinical Experimental Histories—Table 2.

Dog 24-59. Born 1922. Uneventful anemia history 1923 to 1932.

Apr. 28, 1931. *Globin* injection by vein 1 gm. dissolved in 150 cc. 10 per cent dextrose solution. Following the first injection there was a chill but no obvious clinical reaction to subsequent injections.

Dog 27-238. Born Feb., 1927. Continuous anemia history Nov., 1928, to date. Diet at no time contained potent animal protein substances in effort to produce dietary anemia. Experiments consisted of testing vegetables, minerals, drugs and amino acids.

Apr., 1933. One dose of horse globin, 1 gm. in 10 per cent dextrose by vein was followed by shock. Experiment discontinued.

Nov. 18, 1933. Feeding experiment of horse globin, 7.5 gm. daily added to basal bread ration. (See Table 5.)

Feb., 1936. Horse globin by vein, first dose 0.25 gm. in 10 per cent dextrose. Dose gradually increased to 1 gm. which was followed by slight reaction. With dose of 1.25 gm. marked reaction. Dog vomited 10 min. after injection, pulse weak, cyanosis, slight diarrhea. This occurred after 6 doses had been given.

Mar. 9, 1936. Slight shock following 1.25 gm. horse globin. Food consumption poor. Horse globin decreased to 0.75 gm. and later again increased to 1 gm. Considerable reaction following 2nd dose of 1 gm. Dosage decreased to 0.50 gm. and no more than 0.75 gm. given until experiment was complete, a total of 25 gm. horse globin.

Globin given by vein is utilized to form new hemoglobin as seen in Table 2, dog 24-59. This experiment in some respects is different

TABLE 2

Globin (Horse) Given by Vein

Positive and Negative Experiments

Diet periods 1 wk. each	Food consumed	Weight	Plasma volume	R.B.C.	Blood Hb. level	Hb. removed, bled
Dog 24-59. Bull, male, adult						
Food, gm. per day	*per cent*	*kg.*	*cc.*	*mil.*	*per cent*	*gm.*
Bread 450, salm. 75, Kl. 30	100	16.9	1073	4.8	51	1.5
*Globin 1.0, br. 450, salm. 75, Kl. 30	100	17.3	1079	5.5	53	14.5
Globin 1.0, br. 450, salm. 75, Kl. 30	100	17.7	1124	4.9	48	10.4
Globin 1.0, br. 400, salm. 75, Kl. 30	100	17.8	1164	5.7	48	9.7
Globin 0.6, br. 400, salm. 75, Kl. 30	100	17.9	1174	4.8	49	1.3
Bread 450, salm. 75, Kl. 40	100	17.0	1094	5.7	53	15.6
Bread 450, salm. 75, Kl. 40	100	16.6	1054	5.7	42	25.7
Bread 450, salm. 75, Kl. 40	100	16.6	1085	4.6	49	12.9
Bread 450, salm. 75, Kl. 40	100	16.9	1127	5.6	48	26.8
Basal output 10 gm. Hb. per wk. Total net Hb. output 34 gm. Total Hb. = 116.9						
Dog 27-238. Coach, female, adult						
Bread 350, salm. 75, Kl. 20	100	16.8	974	4.5	47	1.3
*Globin 0.9, br. 350, salm. 75, Kl. 20	99	16.9	939	5.5	54	1.5
Globin 0.8, br. 350, salm. 75, Kl. 20	61	16.6	807	5.3	58	11.6
Globin 0.7, br. 350, salm. 75, Kl. 20	85	16.5	805	4.5	52	12.6
Globin 0.5, br. 350, salm. 75, Kl. 20	81	16.5	970	—	41	1.4
Globin 0.7, br. 350, salm. 75, Kl. 20	94	16.7	867	4.5	47	10.4
Bread 275, salm. 20, Kl. 20	84	16.6	949	4.1	47	1.4
Bread 250, salm. 20, Kl. 20	85	16.2	839	5.2	56	15.5
Bread 200, salm. 20, Kl. 20	91	15.9	871	6.0	44	24.4
Bread 275, salm. 20, Kl. 20	95	16.1	976	4.0	53	4.6
Bread 225, salm. 20, Kl. 20	78	16.0	953	4.4	47	1.4
Basal output 9 gm. Hb. per wk. Total net Hb. output 0. Total Hb. = 84.8						

* Total globin given 25 gm.

from a similar experiment given in Table 1. The total globin given was 25 gm. but the injections were spread over 4 weeks and consequently the control after period was likewise 4 weeks. There was no increase in the hemoglobin production (actually a slight decrease)

during the period of injection but the new hemoglobin production appears in the subsequent control weeks. It may not be fair to include the hemoglobin output during the 4th control week and under such circumstances the net hemoglobin output would be 17 gm. instead of 34 gm. as given. At any rate the true hemoglobin return from the injected globin will approximate 100 per cent if we assume that the true net hemoglobin figure is somewhere between 17 and 34 gm. hemoglobin. The toxic influence of the injected globin solution *delayed* the hemoglobin regeneration but did not prevent the body from utilizing the globin material in subsequent weeks.

Table 2, dog 27-238, shows a negative response to globin injections. There is no excess of hemoglobin produced above the basal level and there is some inhibition of hemoglobin production during the prolonged injection period. This dog (see Clinical experimental history 27-238) was quite sensitive to globin injections which produced clinical shock on several occasions and caused lack of appetite. We believe the lack of response to the globin injection with new hemoglobin production to be due to this obvious intoxication. This dog was given one dose of horse globin in 1933 or 3 years before this experiment and at that time the dog was intoxicated to such a degree that the experiment was discontinued. Whether this dose made the dog even more susceptible to subsequent doses we cannot say.

Clinical Experimental Histories—Table 3.

Dog 33-14. Born Nov., 1932. Uneventful anemia history Dec., 1933, to date.

Mar. 8, 1937. Dog globin by vein beginning with 0.75 gm. in 10 per cent dextrose. Dose gradually increased to 2 gm. daily. Some reaction to 2 gm. dose—rapid pulse, considerable coughing. Dog left food for 2 days. Globin omitted for 2 days. Subsequently 0.50 gm. globin caused similar toxic reaction. Globin omitted for 1 day. Globin then injected beginning with 0.50 gm. and gradually increasing to 1.75 gm. daily. A slight reaction to 1.75 gm. was noted. Globin 1.25 gm. for the remaining injection period. No further clinical disturbance. Dog in good condition at end of experiment.

Dog 30-117. Born Sept., 1930. Uneventful anemia history Oct., 1931, to date.

May, 1936. *Dog* globin by vein beginning with 0.25 gm. in 10 per cent dextrose. Dose gradually increased to 2 gm. daily. No reaction at any time. Food consumption 100 per cent throughout experiment. No hemolysis 2 hours after injection of globin.

TABLE 3

Globin (Dog) Given by Vein

Positive and Negative Experiments

Diet periods 1 wk. each	Food consumed	Weight	Plasma volume	R.B.C.	Blood Hb. level	Hb. removed, bled
Dog 33-14. Coach, female, adult						
Food, gm. per day	*per cent*	*kg.*	*cc.*	*mil.*	*per cent*	*gm.*
Bread 350, salm. 75, Kl. 20	100	14.1	803	4.6	44	1.2
*Globin 0.8, br. 350, salm. 100, Kl. 20	100	14.0	672	5.0	53	1.5
Globin 0.5, br. 200, salm. 150, Kl. 20	75	13.7	748	5.5	45	20.3
Globin 1.0, br. 225, salm. 125, Kl. 20	100	13.6	763	4.6	45	1.4
Globin 0.9, br. 225, salm. 125, Kl. 20	100	13.6	812	4.4	44	1.3
Globin 0.9, br. 225, salm. 125, Kl. 20	100	13.4	777	4.7	48	1.4
Bread 250, salm. 100, Kl. 20	100	13.4	745	4.9	50	25.8
Bread 250, salm. 100, Kl. 20	100	13.4	777	4.7	48	1.4
Bread 250, salm. 100, Kl. 20	100	13.4	653	6.0	49	35.3
Bread 250, salm. 100, Kl. 20	100	13.7	806	4.5	45	12.9
Bread 250, salm. 100, Kl. 20	100	13.3	794	5.1	47	24.6
Bread 250, salm. 100, Kl. 20	100	13.4	722	4.5	47	1.4
Basal output 9 gm. Hb. per week. Total net Hb. output 25.0 gm. Total Hb. = 127.3						
Dog 30-117. Coach, male, adult						
Bread 450, salm. 75, Kl. 20	100	15.8	970	4.4	43	1.2
†Globin 0.3, br. 450, salm. 75, Kl. 20	100	16.0	886	5.9	44	22.4
Globin 1.1, br. 450, salm. 75, Kl. 20	100	16.1	944	4.1	39	1.2
Globin 1.6, br. 450, salm. 75, Kl. 20	100	16.0	970	4.1	46	1.3
Globin 1.1, br. 450, salm. 75, Kl. 20	100	16.4	1006	—	36	1.0
Bread 450, salm. 50, Kl. 20	100	16.6	1018	4.5	56	12.2
Bread 450, salm. 50, Kl. 20	100	16.5	1000	5.9	48	20.4
Bread 450, salm. 50, Kl. 20	100	16.4	1025	5.7	43	29.6
Bread 450, salm. 50, Kl. 20	100	16.4	1010	4.1	42	14.5
Bread 450, salm. 50, Kl. 20	100	16.4	1022	4.0	42	1.2
Basal output 14 gm. Hb. per wk. Total net Hb. output 0. Total Hb. = 103.8						

* Total globin given 28.3 gm.

† Total globin given 29 gm.

Table 3, dog 33-14, shows a complete experiment in which 28.3 gm. of globin are given intravenously during a 5 week interval and about 25 gm. of hemoglobin are produced over and above the control base line of 9 gm. hemoglobin per week. There is little hemoglobin produced during the 5 week injection period, distinctly less than the control output and the surplus hemoglobin all appears in the 5 week after period. This reaction is very like that recorded (5) for an endometritis. The globin injections caused some clinical disturbance and only small doses could be given. There was some lack of appetite in the 2nd week of globin injection. We believe this intoxication due to globin injections is responsible for the inhibition of hemoglobin production during the injection period but the building materials are not lost so that a surplus hemoglobin production appears when globin injections are discontinued.

Table 3, dog 30-117, shows a complete experiment in which the globin injections caused inhibition of hemoglobin production in the 4 week injection period but there was no surplus produced in the after period of 5 weeks. This dog showed little if any clinical evidence of intoxication and its appetite was not impaired. The effect of this subclinical intoxication is obvious in the injection period and it seems safe to assume some disturbance due to the globin injections which interfered with the normal mechanism of hemoglobin production. This dog has a high basal hemoglobin output of 14 gm. per week so that the inhibition of hemoglobin construction during the 4 weeks of injection amounts to more than 30 gm. hemoglobin.

Clinical Experimental Histories—Table 4.

Dog 35-1. Born Nov., 1934. Uneventful anemia history July, 1936, to date.

Mar. 9, 1937. Dog globin by vein beginning with 0.25 gm. in 10 per cent dextrose. Dose gradually increased to 2.25 gm. daily. With that amount, slight clinical reaction resulted—rapid pulse and coughing. Dog left all food next day. No globin given for 2 days. Injections of globin again started with dose of 0.50 gm. gradually increased to 1.75 gm. without any further disturbance during rest of experimental period. Dog in good condition at end of experiment.

Table 4, dog 35-1, gives the details of a very interesting experiment. The globin injections caused no significant clinical disturbance and a large total of 31 gm. were given during 4 weeks. There is no

evidence of inhibition of hemoglobin production as the hemoglobin surplus is 59 gm. over and above the base line output of 6 gm. hemoglobin per week. In fact there is a surplus of 28 gm. hemoglobin not explained. We are inclined to this explanation. It is well known in pharmacology that some drugs are destructive or injurious in large doses but stimulating in small doses. This dose of globin in this dog may have been of just the proper amount to cause stimulation rather than inhibition.

TABLE 4

Globin (Dog) Given by Vein—31 Gm.

Hemoglobin Surplus Is 59 Gm.

Dog 35-1. Bull, male, adult.

Diet periods 1 wk. each	Food consumed	Weight	Plasma volume	R.B.C.	Blood Hb. level	Hb. removed, bled
Food, gm. per day	*per cent*	*kg.*	*cc.*	*mil.*	*per cent*	*gm.*
Bread 450, salm. 75, Kl. 20	100	19.5	1147	5.7	47	1.3
Globin 1.1, br. 450, salm. 50, Kl. 20	86	19.3	1152	6.1	55	1.6
Globin 0.6, br. 250, salm. 150, Kl. 20	92	18.5	1085	6.0	53	24.2
Globin 1.3, br. 350, salm. 150, Kl. 20	100	18.6	1142	5.5	38	19.3
Globin 1.5, br. 350, salm. 150, Kl. 20	100	18.8	1210	4.3	38	1.5
Bread 400, salm. 75, Kl. 20	100	18.7	1223	4.8	46	1.4
Bread 400, salm. 75, Kl. 20	100	18.7	1068	5.1	49	12.3
Bread 400, salm. 75, Kl. 20	100	18.9	1096	6.3	36	34.8
Bread 400, salm. 75, Kl. 20	100	18.8	1213	5.0	46	1.4
Bread 400, salm. 75, Kl. 20	100	18.9	1033	5.9	47	25.2
Bread 400, salm. 75, Kl. 20	100	18.6	1124	5.0	43	1.4

Basal output 6 gm. Hb. per week. Total net Hb. output 59 gm. Total Hb. = 123.1

Clinical Experimental Histories—Table 5.

Dog 23-1. Born 1922. Uneventful anemia history, Oct., 1924, to Mar., 1936.

Dec., 1933. Horse globin feeding experiment for 2 weeks with daily dose of 7.5 gm. added to the basal salmon bread ration.

Dog 30-121. Born Sept., 1930. Uneventful anemia history, Oct., 1931, to date.

Mar. 3, 1936. Globin feeding experiment for 2 weeks with daily dose of 7.5 gm. added to basal salmon bread ration.

TABLE 5

Globin (Horse) Given by Mouth

Approximately 30 Per Cent Utilization

Diet periods 1 wk. each	Food consumed	Weight	Plasma volume	R.B.C.	Blood Hb. level	Hb. removed, bled
Dog 27-238. Coach, female, adult						
Food, gm. per day	*per cent*	*kg.*	*cc.*	*mil.*	*per cent*	*gm.*
Bread 350, salm. 125, Kl. 50	100	14.1	910	—	44	1.2
Globin 7.5, br. 375, salm. 100, Kl. 40	100	14.4	847	4.2	40	13.0
Globin 7.5, br. 375, salm. 100, Kl. 40	100	14.7	862	5.0	45	15.4
Bread 375, salm. 100, Kl. 40	100	14.8	868	4.5	48	1.4
Bread 375, salm. 100, Kl. 40	100	14.8	820	6.1	55	37.2
Bread 375, salm. 100, Kl. 40	100	15.1	902	4.7	44	12.9
Basal output 8 gm. Hb. per week. Total net Hb. output 40 gm. Total Hb. = 79.9						
Dog 23-1. Bull, male, adult						
Bread 375, salm. 100, Kl. 40	100	17.9	1099	3.9	39	1.1
Globin 7.5, br. 375, salm. 100, Kl. 40	100	17.8	1063	5.1	51	12.6
Globin 7.5, br. 375, salm. 100, Kl. 40	100	18.0	1125	4.3	45	1.3
Bread 375, salm. 100, Kl. 40	100	17.7	1023	5.3	48	24.7
Bread 400, salm. 100, Kl. 40	93	17.7	1086	4.7	42	11.1
Bread 375, salm. 150, Kl. 50	60	17.0	1030	4.3	42	1.2
Basal output 8 gm. Hb. per week. Total net Hb. output 14 gm. Total Hb. = 50.9						
Dog 30-121. Coach, male, adult						
Bread 450, salmon 50, Kl. 20	100	14.5	878	4.4	43	1.2
Globin 7.5, br. 450, salm. 50, Kl. 20	100	14.7	934	5.3	44	13.2
Globin 7.5, br. 450, salm. 50, Kl. 20	100	14.7	905	5.0	52	14.4
Bread 450, salm. 50, Kl. 20	100	14.9	943	5.8	45	24.6
Bread 450, salm. 50, Kl. 20	100	14.7	904	5.3	50	13.9
Bread 450, salm. 50, Kl. 20	100	14.9	914	4.6	48	1.3
Basal output 9 gm. Hb. per week. Total net Hb. output 27 gm. Total Hb. = 67.4						

Table 5 shows three more experiments with horse *globin feeding*. The first experiment (dog 27-238) is practically a replica of that given in Table 1. A slightly larger amount of globin is fed (total 105 gm. during the 2 weeks period) and a net hemoglobin output of 40 gm. is recorded. The ratio of intake of globin to new hemoglobin production is 105 to 40 or a 39 per cent utilization of this protein to form hemoglobin. This is practically identical to the 40 per cent utilization given in Table 1 and represents maximal utilization for protein given by mouth.

The second experiment (Table 5, dog 23-1) shows a much lower level of protein utilization. With 105 gm. globin given by mouth the net hemoglobin production is only 14 gm.—a 13 per cent utilization. This is about the same as the utilization of liver protein for hemoglobin regeneration—a ratio of 8 gm. intake to 1 gm. hemoglobin output.

The third experiment (Table 5, dog 30-121) shows an intermediate level of protein utilization. Given 105 gm. globin by mouth, the dog produces a surplus of 27 gm. new hemoglobin—a 26 per cent utilization of this protein. The average of all four globin feeding experiments is a 30 per cent utilization—that is given 100 gm. globin by mouth we should expect a net surplus of new hemoglobin amounting to 30 gm.

SUMMARY

It has been shown that the standard anemic dog can use sheep, goose or dog hemoglobin when given by vein and return quantitatively its equivalent as new dog hemoglobin within the red cells.

Globin at times can be used when given by vein with a quantitative return of new hemoglobin in red cells in these same anemic dogs. Again the administration of globin by vein will inhibit the expected hemoglobin formation; and we believe the toxic effect of the globin is responsible. A digest of globin may be used by the anemic dog to form new hemoglobin. Globin from both horse and dog have been tested and seem to react in identical fashion.

The globin radicle of hemoglobin appears to be an important limiting factor in abundant hemoglobin building in this type of anemia due to blood loss.

Globin fed by mouth is well utilized to form new hemoglobin and we may record a 30 to 40 per cent utilization or a return of 30 to 40 gm. new hemoglobin from the feeding of 100 gm. globin. This is to be compared with the utilization of liver protein—an average return of 13 gm. new hemoglobin for the feeding of 100 gm. liver protein.

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