

Humoral Control of Erythropoietic Activity in Man During and After Altitude Exposure.* (29331)

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(Introduced by Alberto Hurtado)

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It is generally accepted that hypoxia is the main stimulus for production of erythropoietine. However, little attention has been given to this factor in relationship to acute and chronic exposure of man to high altitude(1, 2,3). On the other hand, when polycythemic natives of high altitude are brought down to sea level, a decrease of red blood cells occurs. Could this phenomenon be explained on the basis of a diminution of erythropoietine secretion only or is there a humoral inhibitory factor that produces a depression of red cell formation? This report concerns studies which tend to clarify these problems.

Material and methods. For assay of erythropoietine during high altitude exposure, 3 groups of subjects were studied: a) natives of Morococha, at 4,540 meters of altitude; b) newcomers to Morococha after 24 hours, 72 hours and after 8 days of exposure; c) sea level subjects. The plasma obtained from each group was processed following the method of Borsook(4) and lyophilized. This material was tested in albino rats. Each animal received doses equivalent to 4 cc of the original volume of plasma (after 2 days' starvation) on each of 2 successive days; 24 hours after the last injection, 1 microcurie of Fe-59 was given intraperitoneally, and 18 hours later, blood samples from the heart were obtained. Red cell iron incorporation was determined.

The possibility of the existence of an inhibitory factor of the erythropoiesis was studied by testing in rats the plasma filtrate of high altitude natives brought down to sea level after various periods of time: 24 hours, 72 hours, 10 days, and 20 days. The plasma filtrate was prepared following Borsook's

method(4). The rats were previously stimulated by exposing them to a simulated altitude of 18,000 feet for 24 hours. Then 1 cc of the material equivalent to 2 cc of the original volume of plasma was injected intraperitoneally. At the same time, 1 microcurie of Fe-59 was given in the same way. A second injection of plasma filtrate was given 4 hours later; 24 hours after the radioactive iron injection, blood samples were obtained from the heart.

In a second series of experiments, rats were injected with filtrate of plasma from high altitude natives 10 days after their arrival at sea level. One cc of this material equivalent to 4 cc of the original volume of plasma was given for 7 days to one group of rats and for 14 days to a second group. Hemoglobin determinations were done before and after this treatment. The same type of observation was carried out in rats receiving either plasma filtrates of normal sea level subjects or saline.

Results and discussion. An increase of red cell iron incorporation was found only in the group of rats that were injected with plasma filtrates of newcomers to high altitudes after 24 hours of exposure, as compared with the group receiving sea level plasma filtrate (Table I).

These findings are similar to those observed by Stohlman(5) in rats exposed to simulated altitudes, and are also in agreement with the concept that erythropoietine is utilized by the erythropoietic tissue and that it increases in the circulating blood for a few hours only when there is great production.

Most remarkable were the results of tests for a possible inhibitor of the erythropoiesis. Assay animals were exposed to a simulated altitude of 18,000 feet. The animals were given filtrates prepared from plasma of polycythemic subjects of Morococha at intervals following their arrival at sea level. Filtrates

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TABLE I. Plasma Erythropoietic Factor During Exposure to High Altitude.

	Control at sea level	After 24 hr	After 72 hr	After 10 days	Natives of altitudes
Mean $\pm$ S.E.	$5.5 \pm .38$	9.0 ± 1.3	5.5 ± 1.4	5.4 ± 1.2	4.2 ± 1.2
No. of rats	8	7	5	8	7

TABLE II. Effect of Plasma Filtrate from Altitude Natives Brought Down to Sea Level in Rats Subjected to 18,000 Feet.

Group	No. of rats	Treatment Fe-59	Uptake
I	13	Exposed to 18,000 ft, sea level plasma	41.6 ± 2.76
II	7	" " " " 24 hr plasma	35.3 ± 3.48
III	7	" " " " 72 hr plasma	$30.1 \pm 4.04^*$
IV	14	" " " " 10 days plasma	$28.5 \pm 2.46^\dagger$
V	7	" " " " 20 days plasma	$28.4 \pm 2.50^\dagger$

* $P < .05$ $^\dagger P < .01$

TABLE III. Effect of Plasma Filtrate from Altitude Natives After 10 Days at Sea Level, Plasma Filtrate of Sea Level Subjects and Saline on Hemoglobin of Normal Rats.

	No. of rats	Days of treatment	Hb, g % (mean $\pm$ S.E.)		Mean difference	P value	Reticulo-cytes (%)*
			Before treatment	After treatment			
Plasma of altitude natives	5	7	$14.0 \pm .25$	$13.5 \pm .52$	-1.5	$< .05$	$2.6 \pm .63$
after 10 days at sea level	5	14	$16.2 \pm .14$	$12.7 \pm .99$	-3.5	$< .01$	$3.1 \pm .85$
Plasma of sea level sub-jects	5	7	$15.5 \pm .21$	$15.6 \pm .18$	+ .1		$1.9 \pm .45$
	5	14	$15.6 \pm .22$	$15.3 \pm .42$	- .3		$2.0 \pm .50$
Saline	5	7	$15.9 \pm .19$	$16.1 \pm .26$	+ .2		$1.7 \pm .48$
	5	14	$16.1 \pm .17$	$16.4 \pm .23$	+ .3		$2.1 \pm .55$

* Reticulocyte counts were done 18 hr after last injection.

from plasma obtained 72 hours or later caused a significant depression of erythropoiesis (Table II).

In the groups of rats which were given daily injection for a week of plasma filtrates from high altitude natives 10 days after arrival at sea level, the differences of the mean values of hemoglobin before and after the experiments can be seen in Table III.

Both experiments seem to indicate that an inhibitory humoral factor exists in the plasma of high altitude natives brought down to sea level. The presence of such a factor could explain the depression of red cell formation which occurs in them. Also it might account for other problems which cannot be explained on the basis of the existence of a stimulating factor only. Krzymowsky and Krzymowsky have described a depressory effect of plasma of sheep made polycythemic by transfusions (6).

Summary. The erythropoietic stimulating factor was studied in natives and newcomers to high altitude (14,900 feet). An increase

was found only in the group of newcomers after 24 hours of exposure.

The possibility of the existence of an erythropoietic depressing factor is suggested from the investigation of a group of high altitude polycythemic subjects brought down to sea level, where they were followed 72 hours, 10 days and 20 days after arrival. Plasma obtained from them at these time periods when injected into rats had a depressing erythropoietic action.

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