globin would correspond to about one milligram of iron. This amount of iron and even a fraction of it would be easily measurable by the method used, if it were in a labile, open-ring compound.

DISCUSSION

In 1939 Lemberg and his associates[14] confirmed "that the iron of sulphemoglobin can be easily detached," but they pointed out that sulfhemoglobin ". . . cannot be a bile pigment-haematin compound." From the present study it becomes perfectly clear that the iron in sulfhemoglobin as it occurs *in vivo,* at least during enterogeneous sulfhemoglobinemia in rabbits, can not be a part of an open-ring compound, since there is no increase whatsoever in labile iron coincident with the occurrence of sulfhemoglobin in the blood.

It is equally clear that the preparation of sulfhemoglobin *in vitro,* even in experiments of very short H_2S-treatment, is accompanied by an increase of the labile iron, in 8 experiments with human blood averaging about 22 per cent. of the calculated sulfhemoglobin iron.

The question which remains undecided is whether in short-time experiments the substance or substances with labile iron are independent compounds or rather intermediates in the formation of sulfhemoglobin. These may be formed by the well-known action upon heme of nascent hydrogen peroxide originating from oxidation of H_2S.[2, 3] The conversion of the presumptive precursor with labile iron into sulfhemoglobin with firm iron may well be by the entry of sulfur into the molecule.[5, 14, 15] The findings of the present study, together with the data in the literature[5, 12, 14, 15] make it most probable that sulfhemoglobin in its final stable form, as best exemplified when it is produced *in vivo,* is a substance with closed ring. If the ring be opened in a first step reaction, it seems highly reasonable that the reclosure is accomplished by a sulfur-containing group; no direct proof of this last suggestion is available so far.[16]

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[14] R. Lemberg, J. W. Legge and W. H. Lockwood, *Biochem. Jour.,* 33: 754–758, 1939.

[15] F. Haurowitz, *Jour. Biol. Chem.,* 137: 771–781, 1941.

[16] This work, assisted by a grant from the *Fonds d'Etudes "Roche,"* was begun in the Institut de Physiologie de l'Université de Lausanne, Switzerland. One of the authors (G.B.) wishes to express his gratitude to Professor A. Fleisch, head of the named institute, for his friendly hospitality and helpful cooperation during the summer of 1938.

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SKIN SENSITIVITY TO HUMAN PLASMA

A RECENT report of a reaction to the transfusion of human plasma[1] contradicts the published opinion[2] that the intravenous administration of plasma is entirely innocuous. Our own experience with group-specific plasma indicates that reactions do occur. We have employed a number of experimental methods to ascertain the cause of these reactions. One of these, skin testing with plasma, has yielded some new and interesting information.

The plasma used in these experiments was syphoned off freshly drawn blood. The erythrocytes were allowed to settle by sedimentation for from 24 to 72 hours at 4–8° C. before the plasma was removed. The tests were made by injecting 0.05 cc of undiluted plasma intradermally on the volar surface of the forearm. The sites of injection were observed after 10, 30 and 60 minutes.

Positive reactions were indicated by the formation of a wheal 0.8–3 cm in diameter, plus a surrounding zone of erythema. Erythema alone was found to be of no significance. The wheal usually appeared in 10 minutes, reached a maximum in 30 minutes and began to fade at the end of one hour. Citrate and sulfanilamide as used in the preservative and anticoagulant solution yielded uniformly negative skin tests. Of 109 patients tested with the plasma from all blood groups, 20 per cent. were positive to one or more plasmas. Sensitivity to the various plasmas was not confined to individuals of any particular blood group. The positive reactions were not associated with any specific plasma, since the same specimen gave negative and positive results in different individuals. We were able to transfer passively the plasma sensitivity to non-sensitive individuals.

It was possible to transfuse nine of the sensitive patients. Seven of these gave a reaction to the intravenous administration of the same plasma giving a positive skin test. Reactions encountered in the sensitive individuals included headache, dyspnea, epigastric distress, chills, fever and urticaria. No fatal reactions occurred. Plasmas giving negative skin tests have not produced any transfusion reactions.

There are three likely causes for the positive skin tests: allergins, iso-antibodies or A and B substance in the plasma. Schiff[3] has demonstrated the presence of the A factor in serum. We have confirmed the work of Aubert and his coworkers[4] in establishing the presence of A and B substance in plasma. Of the three causes of positive skin tests, it is our belief that

[1] *Jour. Am. Med. Asn.,* 118: 1050, 1942.

[2] *Ann. of Surgery,* 111: 623, 1940.

[3] *Klin. Wschr.,* 3: 679, 1924.

[4] *Jour. Path. and Bact.,* 54: 89, 1942.

the A and B factors are most important in accounting for the dermal reactions. This belief is based on the fact that individuals who show skin sensitivity to A or B plasma are also sensitive to AB plasma. Also individuals sensitive to A or B plasma are sensitive to Witebsky's purified A and B substance.[5]

The correlation between the skin tests and the transfusion reactions is suggestive, but further work is necessary before any conclusion can be reached as to the cause of the reactions. It is not known what pooling of plasmas will do toward the elimination of transfusion reactions. Pooling of plasmas from bloods of incompatible groups failed to neutralize the skin reacting substance. For example, an individual of group O, sensitive to A plasma, still gave a positive skin test after the A plasma had been mixed with an equal volume of B plasma.

From our results it is obvious that plasma reactions do occur, and that there is a correlation between skin sensitivity to the plasma and reactions after plasma transfusions. Negative skin tests preclude the possibility of reactions to intravenous administration of the plasma. Therefore, to prevent reactions, skin tests should be used wherever possible prior to a transfusion.

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SCIENTIFIC APPARATUS AND LABORATORY METHODS

ELIMINATION OF CONTAMINANTS WITH ULTRA-VIOLET RADIATION

SMITH[1] has described an air filtration apparatus with which it is possible to reduce considerably the population of spores of fungi and of bacteria present in the laboratory atmosphere. The purpose of reducing the atmospheric spore load is to enable the preparation and transfer of cultures without losses from contamination. A means of establishing atmospheric sterility is particularly desirable when the conditions are such that an excessive amount of contamination still occurs after every precautionary measure has been employed.

An inexpensive, ultra-violet generator[2] has been devised which is intended for use under laboratory and industrial conditions where the strictest sanitation is desired. A brief study on the practical effectiveness of the generator in reducing air-borne contaminants has indicated that it is superior to the air filtration apparatus described by Smith.

The tests were carried out in a small room in which two 30″ ultra-violet generators are mounted on the ceiling equidistant from the ends of the room. The room is used almost daily as an inoculation chamber for the manufacture of grain spawn. Grain spawn is made by inoculating a sterile rye grain medium in a milk bottle with the mycelium of the cultivated mushroom, *Agaricus campestris*. Mold spores or bacteria which invade the bottle at the time of inoculation grow luxuriantly on the rye grain medium and render the bottle worthless as spawn. In commercial practice a large number of bottles are inoculated at one time. The number of bottles which become contaminated is dependent on the density of the spore population in the atmosphere of the chamber. The air filtration apparatus mentioned above is also mounted for permanent use in the chamber so that an opportunity has been afforded to directly compare the effectiveness of air filtration with ultra-violet radiation in reducing contamination.

In a preliminary test the chamber was contaminated as much as possible by blowing in air from an adjoining unclean room. The door was then closed and ten sterile petri dishes containing potato-dextrose agar were placed on small tables in various parts of the room. These were then exposed to the atmosphere for three minutes each. During the exposure the air in the chamber was kept in constant motion by turning on the air filtration apparatus after closing the air outlet vent. The chamber was again contaminated and the ultra-violet generators turned on. At the end of each hour, for the next five hours, ten petri dishes were exposed in the same manner as the first ten. All the dishes were then incubated for five days. The number of contaminants in each dish was recorded. No distinction was made between a bacterial and a fungus colony. The results are shown in Table 1.

For all practical purposes the atmosphere of the chamber was rendered sterile after 4 hours of radiation.

In another preliminary test 3 common molds which frequently contaminated the bottles were isolated and their lethal dosages determined. A spore suspension was made of an unidentified Penicillium species, an unidentified Alternaria and *Monilia sitophila*. The suspensions were then sprayed over the surface of glass slides covered with a film of agar. The slides were then exposed to the radiation at a distance of 5′ from the source. All the Penicillium spores were

[5] Supplied by Eli Lilly and Company.

[1] N. R. Smith, SCIENCE, 75: 199–200, 1932.

[2] Westinghouse Sterilamp. The radiation is produced by a discharge through a mixture of inert gases in a tube containing mercury vapor at low pressure.