Tableau II. Pourcentages de DOP dans les fragments Fab et Fc de deux protéines de personnes atteintes de la maladie de KAHLER

	Fragment Fab	Fragment Fc
Protéines S (Malade Sen...)	16,4	55,2
Protéines C (Malade Cap...)	8	54,8

DOP (Intersmat). Une ultime vérification est effectuée par couplage chromatographie en phase gazeuse–spectrométrie de masse (spectromètre de masse Finnigan) d'abord en ionisation électronique classique et enfin par réaction ion-molécule en ionisation chimique.

Résultats et discussion. Les acides gras identifiés sont les acides gras habituels des constituants biochimiques humains. Le fragment Fab contient de plus nombreux et de plus abondants acides gras que le fragment Fc, ce qui laisse penser que les acides gras seraient préférentiellement adsorbés sur les chaînes légères de l'IgG[8]. Le DOP apparaît sur le chromatogramme après les acides gras avec un temps de rétention supérieur à 1 h. Sa masse moléculaire, déterminée par ionisation chimique est 390. Nous avons ultérieurement démontré[4], comme SATO[9], qu'il ne s'agissait pas d'une souillure ou d'un artefact.

Le DOP est présent aussi bien dans les IgG normales que dans les IgG myélomateuses. Toutes les IgG myélomateuses ne contiennent pas de DOP, mais nous voyons, dans le Tableau I, que, lorsque ces IgG en renferment, les teneurs en ce phtalate sont plus élevées que dans les IgG normales.

Nous avons essayé, en particulier sur les sérums myélomateux de deux malades, de mettre en évidence la localisation principale du DOP sur l'IgG. Il apparaît très nettement, dans le Tableau II, que le DOP est lié au fragment Fc, ce qui peut permettre de dire qu'il est préférentiellement attaché aux chaînes lourdes de l'IgG.

Nous avons dit plus haut que le DOP est un plastifiant. Sa liposolubilité a déjà été signalée par MARCEL et NOEL[10]. WESSMAN[11] a démontré qu'un autre phtalate, le diéthylhexyl-phtalate (DEHP), était susceptible de contaminer les protéines du sérum et en particulier les IgG, les transfusions apportant aux malades des quantités substantielles de DEHP.

Dans le Tableau I, nous constatons que, sauf dans les stérides, les pourcentages de DOP trouvé dans un sérum normal conservé dans des «vacutainers» en verre, sont inférieurs à ceux de DOP contenu dans des IgG normales et myélomateuses.

SMITH[12] et TURNER[13] ayant démontré la toxicité, à dose élevée, des plastifiants, la généralisation, en milieu hospitalier, du matériel à usage unique en matière plastique pourrait poser des problèmes.

Nous recherchons actuellement différentes voies de contamination par les phtalates telles que le sang transfusé et stocké dans des récipients en PVC et l'ingestion d'aliments[9] contenus dans des emballages en matière plastique.

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Auto-Oxidation of Hemoglobin in Plasma

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Summary. The rate of auto-oxidation of human hemoglobin to methemoglobin was measured in plasma at 37 °C. Half-lives of hemoglobin were found to be 20 h, 12 h and 7 h at the oxygen tensions of 126, 57 and 23 mm Hg, respectively.

Hemoglobin, which is released from erythrocytes into plasma during an episode of intravascular hemolysis, will rapidly bind to a specific protein, called haptoglobin, and the resulting complex will be taken up by the liver[1]. When the binding capacity of haptoglobin is exhausted, free hemoglobin will either be cleared through the kidney[2], or it will be irreversibly oxidized[3,4]. The resulting methemoglobin readily dissociates, and its ferriheme groups will be attached to two serum proteins -- hemopexin and albumin[5].

It has long been known that hemoglobin is slowly oxidized in solutions containing dissolved oxygen, and that the rate of oxidation is enhanced by low pH[6,7], high ionic strength[6], organic phosphates[8] and low oxygen pressure[8,9]. The purpose of this study was to re-examine the rate of oxidation at different tensions of oxygen, but under conditions resembling those in circulating plasma.

Materials and methods. Oxyhemoglobin was prepared from hemolysates of stored adult human blood[10], and dialyzed against several changes of 0.9% NaCl (final concentration 100 mg/ml). This solution was added to a normal CPD-plasma at a concentration of 1000 mg/100 ml (pH 7.25 at 37 °C, P_{CO_2} 41 mm Hg). Incubations were performed in rotating tonometers at 37 °C[11]. Gas mixtures contained 3.5, 8.5 and 19.0% oxygen, in addition to

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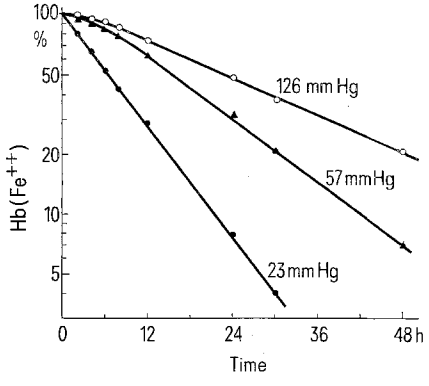
6.2% CO_2 and nitrogen, corresponding to oxygen tensions of 23, 57 and 126 mm Hg, respectively. Aliquots were sampled at fixed intervals for analysis. Oxygen content of the solution was determined by gas chromatography[12], and the O_2-saturation of hemoglobin was computed. For methemoglobin determination, calibration curves were constructed from absorbances of known mixtures of oxyhemoglobin, methemoglobin and plasma, which were pre-incubated for 1 h at room temperature. Identical results were obtained with two calibration curves, using differences A_{577}–A_{505} and A_{577}–A_{630}, respectively.

Results and discussion. Oxygen saturation was determined following 30 min of equilibration with the gas mixture. The resulting oxygen dissociation curve shows a marked shift to the left when compared with the standard dissociation curve of whole human blood: HbO_2 (126 mm) = 99%; HbO_2 (57 mm) = 92%; HbO_2 (23 mm) = 54%. The observed high affinity for oxygen is in accord with results of BUNN et al.[13] who demonstrated a P_{50} of 17 mm Hg under similar conditions. Auto-oxidation rate at 126 mm Hg seems to increase with the time of incubation, reaching a constant value after 12 h (Figure). This time-dependence of oxidation rate can be explained by either inactivation of a protective mechanism or release of an auto-oxidation enhancing principle. Red cell hemolysate and plasma contain a number of substances which are known to affect the oxidation rate of hemoglobin by dissolved oxygen, i.e. organic phosphates[8], copper ions[14], glutathione[15] and the complex enzymatic system for methemoglobin reduction[16]. Further investigations are necessary to recognize the relative importance of these constituents in auto-oxidation of plasma hemoglobin under physiological conditions. Slow production of methemoglobin in the initial period is comparable to that demonstrated by BUNN et al.[13]. Half-life of the fully oxygenated hemoglobin in plasma ($T_{1/2}$ = 20 h) seems to be half as long as that found in dilute buffer solutions[7]. In addition, the auto-oxidation curve in buffer showed a biphasic course, the initial rate being faster than the consecutive[7], a finding at variance with our results with plasma. Half-lives of oxyhemoglobin at 57 and 23 mm Hg were 12 and 7 h, respectively. These values again indicate a considerably faster oxidation of hemoglobin in plasma than in a buffer solution[7]. Provided that an extrapolation of these results is admittable, a rather fast in vivo oxidation of free hemoglobin has to be expected. However, a reliable estimation of the in vivo oxidation rate is difficult, since the oxygen tension of circulating blood fluctuates in the vascular bed. If 60 mm Hg is a reasonable assumption of the mean intravascular O_2-tension, free hemoglobin may undergo auto-oxidation with an approximate half-life of 12 h.



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Effects of Two Kinds of Social Deprivation on Testosterone and Estradiol-17 β Plasma Levels in the Male Rat

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Summary. The effect of complete and tactile isolation on plasma testosterone and estradiol-17 β was studied in male rats at the age of 60 and 180 days. A decrease in the plasma levels of the two hormones was observed.

In recent years many investigations produced evidence that modifications of the social environment can affect endocrine parameters in rodents. It is well established that adrenal[2–7] and gonadal[4, 6, 8–10] functions are involved in the effects produced by social deprivation, and particularly by isolation.

Isolation influences also many behavioural traits, particularly sexual behaviour[11–14]. GERALL et al.[13] and SPEVAK et al.[14] found differences in sexual behaviour between socially reared and isolated rats, but did not find differences between subjects reared in total and in contact isolation.

The purpose of this investigation is to study the modifications of testosterone and estradiol-17 β plasma concentration induced at 60 and 180 days of age by varying degrees of social deprivation in the rearing of the male rat. For this study, 48 Sprague Dawley rats from a stock maintained in our laboratory (originally obtained from Charles River) were used. At the age of 21 days, subjects were weaned and randomly divided into 3 groups of 16 rats each.

Group 1. Total isolates. Animals were raised alone in a cage 15 cm wide by 12 cm high by 33 cm long. They could receive auditory and olfactory stimuli. Group 2. Contact isolates. Animals were separated by a wire mesh screen and so disposed that each subject was adjacent to a male and a female. Each compartment was 25 cm wide by 20 cm high by 25 cm long. Group 3. Socially reared. 4 males and 4 females were housed in a cage (38 cm wide $\times$ 60 cm long $\times$ 20 cm high). Litters delivered in the cage were removed at the age of 20 days.

Eight animals of each group were sacrificed at the age