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Effect of radiographic contrast media (Iodixanol, Iobitridol) on hemolysis

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Abstract. Radiographic contrast media are able to induce changes of the morphology of erythrocytes and endothelial cells. Particularly, the change of the erythrocyte morphology is associated with a decreased deformability possibly resulting from disintegration and a loss of constituents of the membrane cytoskeleton.

However, it is unclear whether there is an intravascular hemolysis as a consequence of the disintegration of the erythrocyte membrane cytoskeleton which might more or less coincide with a loss of erythrocyte membrane integrity.

The results of this study showed, that free hemoglobin increased from 14.2 ± 5.1 mg/dl to 17.9 ± 9.8 mg/dl after Iobitridol application ($p = 0.089$), while it slightly decreased from 21.5 ± 10.9 mg/dl to 19.0 ± 12.9 mg/dl after Iodixanol application ($p = 0.289$). The slight decrease of free hemoglobin after application of Iodixanol differed significantly compared to the increase of free hemoglobin after Iobitridol application ($p < 0.05$).

This different reponse is thought to give evidence to the assumption that the erythrocyte membrane integrity, in deed, was compromised leading to the release of free hemoglobin as an indicator of hemolysis as well.

Keywords: Erythrocytes, hemolysis, radiographic contrast media, Iodixanol, Iobitridol

1. Introduction

Radiographic contrast media (RCM) are iodine containing solutions which are widely used for imaging of the vascular system. RCM exhibit great differences in their physicochemical properties e.g. their carboxyl and hydroxyl groups, osmolality and chemotoxicity [6, 7]. Several studies revealed enormous differences in the RCM-induced formation of echinocytes [1, 11, 13, 16, 17, 20–22, 28] and also in the aggregability of erythrocytes [10] which also might influence the microcirculatory blood flow [14, 15]. These differences could be due to variations in the loss of constituents of the membrane cytoskeleton (e.g. Actin), to more or less drastic changes in the conformation of the spectrin network [9] and to a loss of binding elements needed to fix the membrane cytoskeleton to the erythrocyte membrane [9, 12]. The clustering of band3, particularly, is discussed to reduce the deformability [18, 19, 23, 25], to lead to the abridgement of the lifespan and to removal of erythrocytes by offering senescence signals [2].

However, it is not clear whether there is an intravascular hemolysis as a consequence of the demonstrated damage of the red cell membrane. A direct result of hemolysis of RBCs would be the release of hemoglobin into the blood plasma leading to circulation of free hemoglobin.

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Table 1
Demographical and clinical data of both patient groups (CAD: coronary artery disease; HLP: hyperlipoproteinemia)

RCM	Age [years]	male	CAD / myocardial infarction	hypertension	diabetes	HLP
Iodixanol320 ($n = 20$)	73.8 ± 7.9	13	10/2	20	6	15
Iobitridol350 ($n = 20$)	78.1 ± 20.1	14	11/1	17	7	11

Therefore a study was started as a part of a quality management project in the heart catheter laboratory in order to answer the question if Iobitridol350 in comparison to Iodixanol320 could have an influence on the release of free hemoglobin during a diagnostic coronary angiography.

2. Material and methods

The study was performed as a part of a quality management project in the heart catheter laboratory in compliance with the Declaration of Helsinki/Somerset West [24]. The patients received radiographic media (Iobitridol 350 or Iodixanol 320) during diagnostic heart catheter angiography.

The $n = 20$ patients enrolled in each group exhibited typical cardiovascular risk profiles (see Table 1). In each group merely two patients were acute smokers. Both groups did not differ with respect to the outlined variables.

3. Results

Table 2 shows that the glomerular filtration rate (GFR, calculated according to the MDRD formula), the left ventricular function, the total amount of RCM applied and the concentration of free hemoglobin did not differ in both patient groups before the coronary angiography (two-sided *t*-test for unpaired samples: $p > 0.05$ each).

Whereas free hemoglobin increased from 14.2 ± 5.1 mg/dl to 17.9 ± 9.8 mg/dl after Iobitridol application ($p = 0.089$), it decreased from 21.5 ± 10.9 mg/dl to 19.0 ± 12.9 mg/dl after Iodixanol application ($p = 0.289$). The decrease of free hemoglobin after application of Iodixanol ($+2.75 \pm 8.9$ mg/dl) has shown significant differences compared to the increase of free haemoglobin after Iobitridol (-3.9 ± 10.2 mg/dl) application (two-sided *t*-test for un-paired samples: $p = 0.044$).

4. Discussion

The study revealed that the two RCM used in this study affected the erythrocyte membrane in a different way possibly by influencing the erythrocyte membrane integrity. After Iobitridol application

Table 2
Glomerular filtration rate (GFR), left ventricular function (LVEF), total amount of RCM applied (RCM volume) and the concentration of free hemoglobin (fHb) before and after coronary angiography in patients suspected to suffer from coronary artery disease

RCM	GFR [ml/min]	LVEF [%]	RCM-volume [ml]	fHb [mg/dl] befre Angio	after Angio
Iodixanol ($n = 20$)	56.5 ± 6.7	47.5 ± 13.8	104.9 ± 9.5	21.5 ± 10.9	19.0 ± 12.9
Iobitridol ($n = 20$)	60.2 ± 1.8	44.1 ± 7.8	99.9 ± 0.71	14.2 ± 5.1	17.9 ± 9.8

an intravascular increase of the concentration of free hemoglobin happened in spite of the increase in blood volume due to a shift of interstitial fluid into the vasculature caused by the hyperosmolality of Iobitridol (915 mOsmol/(kg H2O)) [5]. This probably led to a certain dilution of free haemoglobin and to lower values of free hemoglobin concentrations, so that the effect might even be underestimated. The intravascular application of hyperosmolar RCM after a prior hydration therapy always carries the risk of an acute hypervolume load for patients suffering from heart insufficiency and from a reduction of kidney function further aggravated by RCM molecule elimination [3, 26].

Iodixanol is isoosmolar and the application leaves the intravascular blood volume practically unaltered. The slight decrease of free haemoglobin after Iodixanol application is thought to be a consequence of the infusion of electrolyte solution (and is in the range of the measuring precision). Equivalent volumes of electrolyte solutions accompanied the application of Iobitridol as well as of Iodixanol. One patient in each of both groups (both suffered from diabetes mellitus and from reduced kidney function) received 1 l of electrolyte solution also before coronary angiography.

The increase of free hemoglobin after Iobitridol application is thought to give evidence to the assumption that the erythrocyte membrane integrity was compromised leading to the release of free hemoglobin indicating hemolysis. One possible mechanism responsible for hemolysis might be the induction of pores in the erythrocyte membrane as in the case of effective membrane attack complexes after complement activation [4]. An *in vitro* study on the influence of RCM on human erythrocytes revealed that RCM application can imply an activation of the complement system. Also, after the application of hyperosmolar RCM in patients with coronary artery disease, plasma complement marker levels were markedly stronger increased ($29.3 \pm 5.5\%$) than after the application of the isoosmolar RCM Iodixanol ($0.47 \pm 0.70\%$) [8]; this difference was significant ($p < 0.00001$). In a second study in healthy volunteers Visipaque induced no or only a weak activation of the complement cascade [27].

5. Conclusion

After Iodixanol application the concentration of free hemoglobin did not change in patients. In contrast, after application of Iobitridol an increase of free hemoglobin was ascertained. This is thought to give evidence to the assumption that the erythrocyte membrane integrity was compromised leading to the release of free hemoglobin as an indicator of RCM-induced hemolytic activity.

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