

BACKGROUND

The CSMR (Centro Studi Microcitemie di Roma) deals from more than 50 years with prevention and diagnosis of Thalassaemia syndromes and haemoglobinopathies. The detection of the haemoglobin fractions HbA₂ and HbF, very important parameters to reach diagnostic results, is currently performed using the cation exchange HPLC method. In this study we compared the results obtained for the haemoglobin fractions HbA₂ and HbF by the Variant II system (BIO-RAD Laboratories, Hercules, CA, USA) and the G8 system (TOSOH HLC-723 G8). Furthermore, when possible, we compared the analytic results of abnormal haemoglobin fractions according to identifying and quantitative capacity.

METHODS

156 samples coming from our ambulatory were studied. These samples were analyzed as routine, by the HPLC Variant II (BIO-RAD Laboratories, Hercules, CA, USA) and successively, they were examined by the G8 system (TOSOH HLC-723 G8). The analyzed samples were divided into three subgroups:

(a) HbA₂ < 2.0% (n=15) (b) 2.1% < HbA₂ < 3.2% (n=111) (c) HbA₂ > 3.3% (n=30)

In all subgroups the median calculation was carried out for the HbA₂ and HbF.

Table I. HbA ₂ median						Table II. HbF median					
HbA ₂ < 2.0%		2.1% < HbA ₂ < 3.2%		HbA ₂ > 3.3%		HbF < 1.1.0%		1.2% < HbF < 2.0%		HbF > 2.1%	
TOSOH G8	VARIANT 2	TOSOH G8	VARIANT 2	TOSOH G8	VARIANT 2	TOSOH G8	VARIANT 2	TOSOH G8	VARIANT 2	TOSOH G8	VARIANT 2
1,30	1,80	3,00	3,00	5,20	4,35	0,50	0,30	1,30	1,20	3,80	3,60

We used the Pearson correlation coefficient to compare the HbA₂ and HbF obtained values. Some samples already selected by the Variant II for the presence of rare haemoglobin variants, were tested by the G8 as well.

RESULTS

The analysis of the results obtained by the two methods pointed out a correlation coefficient of 0.99 for both HbA₂ and HbF. This result indicates a very strong correlation.

Table III. HbA ₂ values correlation coefficient according to 2 different diagnostic systems			
Total patterns	HbA ₂ <2.0%	2.1% < HbA ₂ <3.2%	HbA ₂ > 3.3%.
TOSOH vs VARIANT	TOSOH vs VARIANT	TOSOH vs VARIANT	TOSOH vs VARIANT
0,99	0,95	0,80	0,95

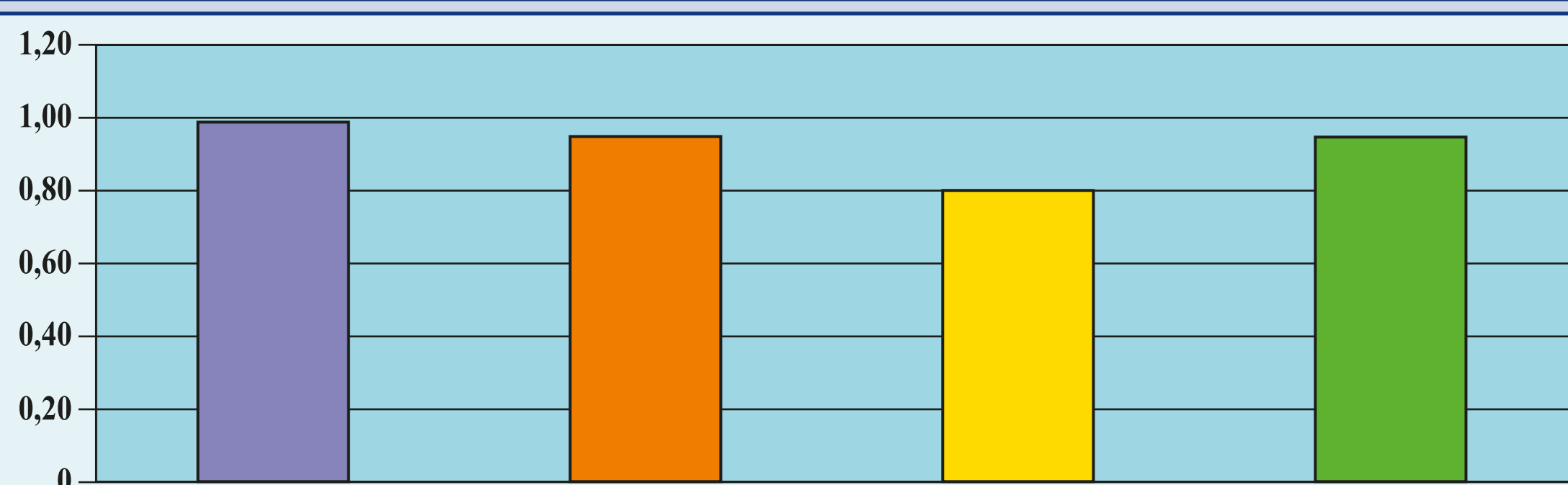


Fig. 1: Graphic representation of the correlation between HbA₂ values according to 2 different diagnostic systems

Table IV. HbF values correlation coefficient according to 2 different diagnostic systems			
Total patterns	HbF < 1.0%	1,2% < HbF < 2,0 %	HbF > 2.1%.
TOSOH vs VARIANT	TOSOH vs VARIANT	TOSOH vs VARIANT	TOSOH vs VARIANT
0,99	0,91	0,87	1,00

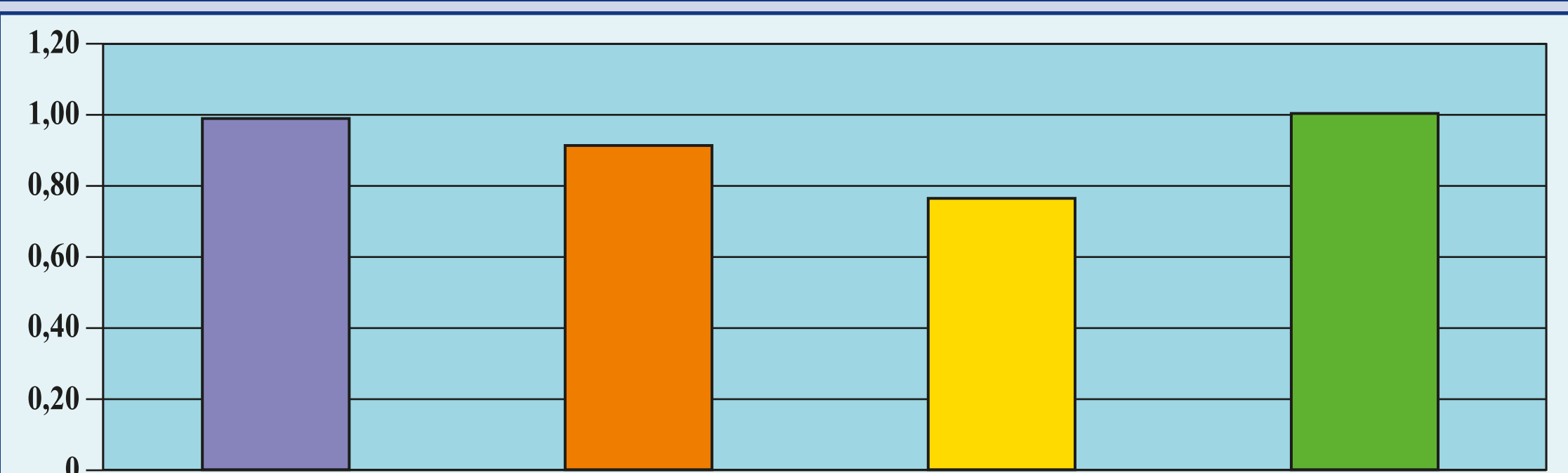
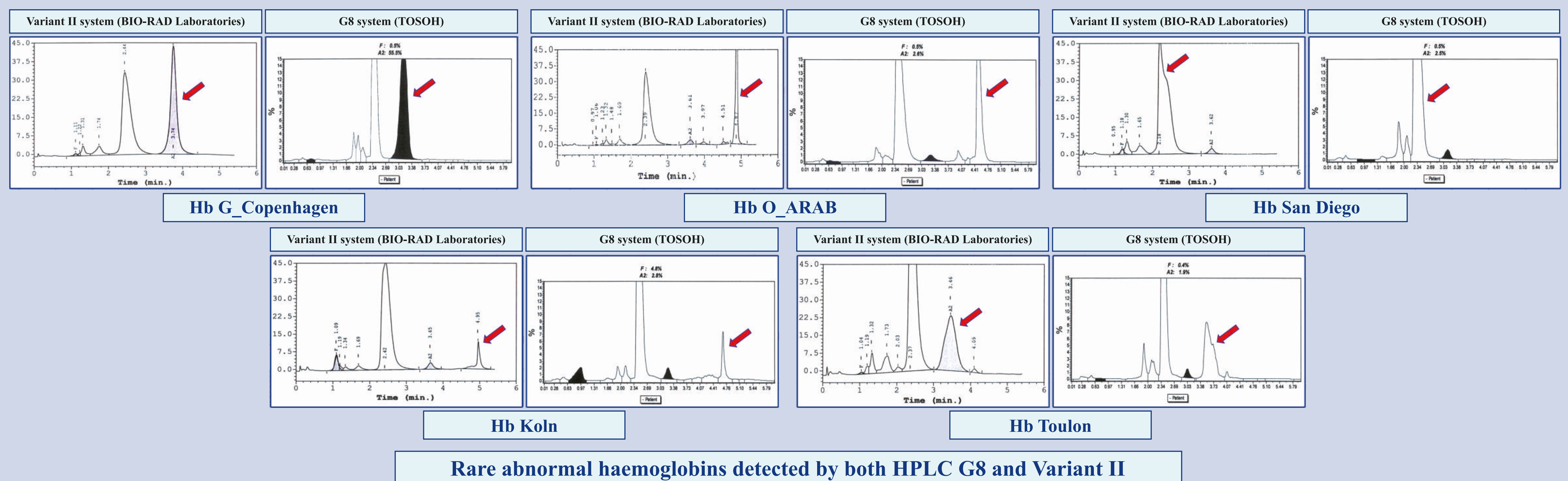


Fig. 2: Graphic representation of the correlation between HbF values according to 2 different diagnostic systems

As for the HPLC separation of the examined haemoglobin variants, we obtained excellent results by the G8 system even if we observed the retention times are different respect with Variant II's. We highlighted the following haemoglobin variants: Hb G Copenhagen, Hb O Arab, Hb San Diego, Hb Koln, Hb Toulon and one more Delta variant still under identification.



CONCLUSIONS

Both systems provided equivalent results that can be interpreted in the same way in diagnostic terms. The excellent overlap obtained by two different systems, leads us to consider the G8 HPLC as a reliable support for first level haemoglobin diagnostics.