[原著]

事前検査におけるヘモグロビン測定の導入

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Implementation of measuring hemoglobin concentration at pre-donation test

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抄　録

香川県赤十字血液センターでは2003年10月に，事前検査として血液比重にかわって，ヘモグロビン(Hb)測定法を導入した。Hb法の最大の利点はその定量性にあり，献血者にHb値を数字として提示することができ，Hb低値者，高値者に対する対応を明確にし得た。また，懸念されていたHb不足による献血不適格者数，VVR発症率も比重法施行時と大差がなかった。今回の検討で，Hb12.5g/dL以上がほぼ比重1.053以上に，12.0g/dL以上が1.052以上に相当すること，Hbと赤血球指数との関係から，赤血球が正色素性から小球性低色素性に変わるHb値が12.5～12.0g/dLであることから現行の採血基準は妥当であると考えられた。Hb法は測定装置がHbの表示まで時間を要すること，温度差による配慮が必要であるなどの欠点はあるが，定量性，均一性を重視するGMPからみても従来の比重法より優れていると結論した。

Key words: Pre-donation examination, Hemoglobin determination
Blood donation criteria, HemoCue hemoglobin analyzer

はじめに

香川県赤十字血液センターでは，2003年10月より，事前検査として硫酸銅法による比重測定にかわって，簡易ヘモグロビン(Hb)測定装置，ヘモキュウヘモグロビンシステム(以下Hb法)による方法に変更した。採血基準は，血液事業の根幹の一つであり，その判定には定量的なHb法が最も妥当と考えられるゆえである。自動血球算定装置がルーチン化したわが国において，貧血の診断はすべてHb，ヘマトクリット，赤血球数によっており，目視による比色法(ザーリ法)や比重法(硫酸銅法)は赤十字血液センターを除いて用いられていない。最近の献血の適否に関する世界の論文は，すべてがHb法を用いて判断しており[1～3]，比重法は

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検査法として教科書の記載すらない現状である[4]。

今回，比重法とHb法の比較，変更前後の献血不適格者の比率，副作用，とくに血管迷走神経反応(Vasovagal Reflex：以下VVR)の比率，また，200mL献血12.0g/dL以上，400mL献血12.5g/dL以上とされている採血基準の妥当性についても検討した。さらに，Hb法の有用性を生かして，不適格者のHb濃度別による個人指導のありかたについても検討したので，これらの成績を報告する。

方　法

簡易Hb法(ヘモキュウ)によるヘモグロビン測定は，あらかじめ試薬が充填された専用マイクロキュベットに10μLの末梢血をサンプリングしアナライザーにセットして，表示されるHb量を読み取る。Hb測定はアザイドメトヘモグロビン法により570nmと880nmからなる2波長様式によっている。

200mL献血申込者63名，400mL献血申込者62名において，血液比重測定と同時に自動血球計数装置(STKS)によるHb測定を行い両法の比較を行った。次に，平成14年4月1日から15年3月31日の間に比重法によって判定した献血者と平成16年4月1日から17年3月31日の間にHb法で判定した献血者において，本社採血基準による献血不適格者の比率，VVRの発症比率を比較検討した。また，献血申込者男性1,472名，女性771名のHb法によるHb濃度別度数分布を作成した。次に，STKSによって得られたMCV，MCH，MCHCとHb値の関係をみることにより，Hb法採用時の採血基準の妥当性を検討した。

Hb法(ヘモキュウ)を導入して1年6カ月経過した時点で，献血バスで実際に使用している看護師17名にアンケート調査を行った。

結　果

1．比重法とHb法の関係

400mL献血申込者のうち，血液比重1.053以上を示した献血者62名のHb値は12.6～17.3g/dLの範囲になり，その平均値±1SDは14.96±1.12g/dLであった。同様に比重1.052以上の200mL献血申込者63名は12.1～16.4の範囲で平均値は13.64±1.16g/dLであった。以上から，400mLの採血基準1.053以上またはHb12.5g/dL以上，200mLの採血基準1.052以上または12.0g/dL以上は両者ともcut off値として妥当であると考えられた。また，比重法の結果はHb値で幅広い範囲に分布し，定量性がないことも明らかとなった。

2．簡易Hb法と自動血球計算装置との相関

簡易Hb法(ヘモキュウ)と自動血球算定装置(Coulter STKS)によって測定した結果の相関を図1に示した。相関係数0.951(Y＝0.8893X＋1.59)の高い相関がみられた。

3．Hb法による献血者ヘモグロビンの度数分布

Hb測定の定量性を生かして献血者ヘモグロビンの度数分布が得られた(図2)。献血申込者の男性1,472名，女性771名の解析で最も頻度が高いのは，男性15.0～15.5g/dL，女性12.5～13.0g/dLであった。

4．比重法およびHb法による献血不適格者の比較

表1に比重法(平成14年4月1日～15年3月31日)とHb法(16年4月1日～17年3月31日)で判定した比重あるいはHb不足による献血不適格者の比率を示す。両者の年齢区分毎不適格率で大きな差異は認めなかった。200mL，400mLの合計において比重法の男性申込者は23,985名，うち不適格者数(率)151名(0.6%)，女性申込者は21,715名，うち不適格者4,404名(20.3%)，Hb法の男性申込者22,749名，不適格者数(率)151(0.6%)，女性申込者20,504名，不適格者数3,958名(19.3%)で，いずれも差異を認めなかった。400mL申込女性で40歳代では，多数の(26～30%)不適格者がみられた。また，400mL申込女性でHb12.5g/dL未満431名のうち10.0g/dL未満が43名(10.0%)，8g/dL未満も4名みられ，治療を必要とすると考えられた。

5．献血時副作用の比較

輸血副作用のうち採血基準が関係すると思われるvaso-vagal reaction(VVR)の発症率を比較した。ヘモキュウが用いられる献血バス200mL，400mL採血のVVRはHb法で男性が減少していたが，女性での頻度の差は認められなかった(表2)。いずれにしてもHb法を導入してVVRが増加することはなかった。

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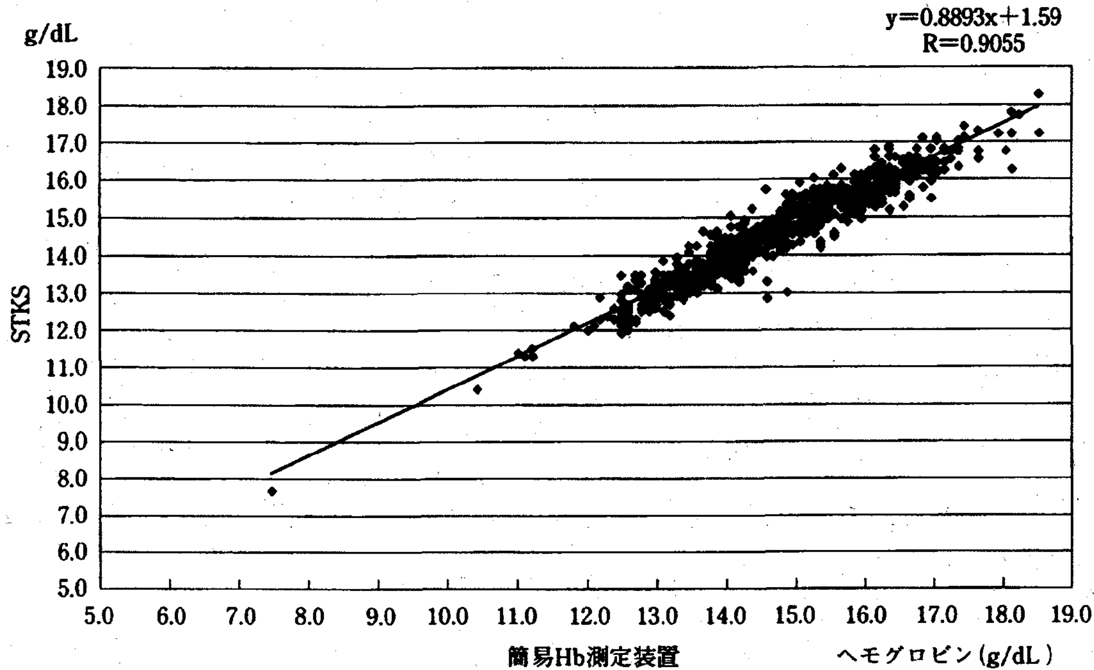
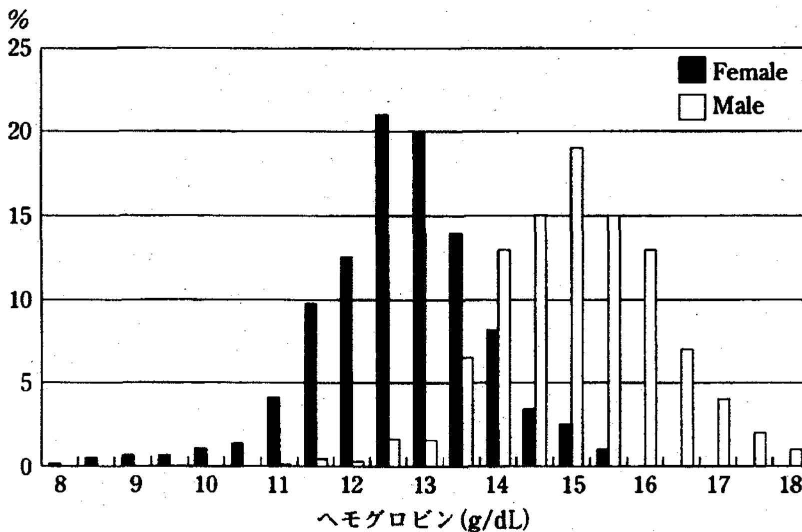


図1　簡易Hb測定装置(ヘモキュウ)と自動血球算定装置(STKS)との比較



献血申込者，男性1,472名、女性771名のヘモグロビン分布。男性で最も多いのは15.0～15.5g/dL，女性で最も多いのは12.5～13.0g/dLであった。

図2　献血申込者のヘモグロビン値の分布

6．ヘモグロビンと赤血球指数の関係

Hb値と赤血球指数(MCV，MCH，MCHC)の平均値の関係を表3に示す。Hbの低下に伴って赤血球指数も低下してくる。低下傾向が認められるのは男性で，MCV，MVH，MCHCともHb12.5g/dL未満から，女性12.0g/dL未満からであり，小球性低色素性の傾向が認められるのは男性が0.5g/dL高かった。以上から，Hbの低下にともなって赤血球は12.5～12.0g/dLで正色素性から小球性低色素性に変わることが判明した。

7．Hb低値による献血不適格者への対応

Hb測定の定量性を生かして献血者のHb値に応じた指導を行うこととした。Hb値10g/dL未満の献血者には医療機関を受診し治療を受けるよう医

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表1　比重法およびHb法による献血不適格者の比較

			年齢区分	19～19	20～29	30～39	40～49	50～59	60～69	計
比重法	男性	200	申込数	1,091	286	346	550	517	210	3,000
			不適数	8	0	5	5	15	1	34
			不適率	0.7	0	1.4	0.9	2.9	0.5	1.1
		400	申込数	1,040	4,464	5,683	5,198	3,659	941	20,985
			不適数	5	14	21	29	30	18	117
			不適率	0.5	0.3	0.4	0.6	0.8	1.9	0.6
	女性	200	申込数	2,240	3,139	2,938	1,976	1,904	689	12,877
			不適数	399	602	689	448	239	67	2,444
			不適率	17.8	19.2	23.5	22.8	12.6	9.7	19.0
		400	申込数	601	1,923	2,097	1,923	1,771	523	8,838
			不適数	110	446	588	582	198	36	1960
			不適率	18.3	23.2	28.0	30.3	11.2	6.9	22.2
Hb法	男性	200	申込数	1,050	298	340	421	448	224	2,781
			不適数	7	1	1	4	5	8	26
			不適率	0.7	0.3	0.3	1.0	1.1	3.6	0.9
		400	申込数	1,147	4,183	5,510	4,832	3,373	923	19,968
			不適数	2	9	17	24	31	18	101
			不適率	0.2	0.2	0.3	0.5	0.9	2.0	0.5
	女性	200	申込数	2,422	2,579	2,825	1,762	1,510	612	11,710
			不適数	461	425	593	386	140	64	2,069
			不適率	19.0	16.5	21.0	21.9	9.3	10.5	17.7
		400	申込数	601	2,038	2,286	1,786	1,584	499	8,794
			不適数	176	454	596	467	163	33	1,889
			不適率	29.3	22.3	26.1	26.1	10.3	6.6	21.5

表2　比重法およびHb法によるVVR発症率の比較

		男　性	女　性
比重法	軽症	83	53
	重症	1	1
	計	84	54
	発症率（%）	0.44	0.43
Hb法	軽症	44	50
	重症	3	2
	計	47	52
	発症率（%）	0.27	0.44

師が指導し，12g/dL未満，10g/dL以上の献血者には食事指導用のパンフレットを作成し配布すると同時に，月に1度栄養士会による個別栄養指導も開設した。

8．Hb高値の献血者の頻度

採血可能であった男性1,472名，女性771名について（図2），Hb17.0g/dL以上の比率は，17.5＞Hb≧17.0：30例（3.0%），18.0＞Hb≧17.5：3例（0.3%），18.5＞Hb≧18.0：3例（0.3%），19.0＞Hb≧18.5：1例（0.1%）の計37例で，いずれも男性で女性にはみられなかった。また，赤血球指数は正常であった。

9．ヘモキュウ使用者のアンケート結果

ヘモキュウを使用している看護師のアンケート結果は以下のとおりであった。まず，利点としては①感染性廃棄物としての後始末が簡単になった（100%），②測定法が簡単である（74%），③献血者にHb値を示すことで説得力がある（63%），などであった。欠点としては①外気温や光線の影響

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表3　Hbと赤血球指数の関係

Hb(g/dL)	男性			女性		
	MCV (fl)	MCH (pg)	MCHC (g/dL)	MCV (fl)	MCH (pg)	MCHC (g/dL)
16.0>Hb≧15.5	93±4	32±2	34±0			
15.5>Hb≧15.0	93±5	32±2	34±1	93±4	32±2	35±1
15.0>Hb≧14.5	92±3	32±2	34±1	92±3	32±1	35±0
14.5>Hb≧14.0	92±5	32±2	34±1	91±3	31±1	35±0
14.0>Hb≧13.5	92±4	32±2	35±1	91±1	32±1	35±0
13.5>Hb≧13.0	92±6	32±2	34±0	90±4	31±2	35±1
13.0>Hb≧12.5	92±5	32±2	34±1	90±3	31±1	34±0
12.5>Hb≧12.0	84±6	28±3	34±1	91±6	31±2	34±0
12.0>Hb≧11.5	83±5	28±2	34±0	87±5	30±2	34±1
11.5>Hb≧11.0*	77±0	25±0	33±0	83±5	28±2	34±0
11.0>Hb≧10.5				83±6	27±2	34±1

n=20 (*n=2)

を受けやすい(94%)，②測定に時間がかかる(94%)，③新たに精度管理が必要になった(69%)，などであった。

考　案

従来から採血基準として用いられている硫酸銅法による血液比重は，献血者を1.052未満，1.052以上(200mL)，1.053以上(400mL)と3区分して可否を判定するもので，各区分内に様々なヘモグロビン濃度が含まれる定性法であり，血液事業が始まって以来半世紀あまりずっと用いられている。しかしながら，比重法は測定者により±0.001程度のバラツキがあることが指摘されている[5]。一般に，赤血球沈降速度は，高温で促進，低温で遅延し補正が必要とされている[6]。佐野らの検討では，10℃で20℃に比し，0.001～0.002低い値，30℃で0.001～0.002高い値が得られるとしている[5]。また，Jamesら[7]は比重法の方がHb法よりも偽の適判定(false-pass)が多いことを証明した。以上から，現在のGMPに準拠した血液事業の理念からすれば，いつ，誰が，どう行っても一定した数値が得られるHb法の方が理想的であることは明白である。今回，簡易ヘモグロビン測定装置(ヘモキュウ)を導入して2年あまりになるので，従来の比重法との比較を様々の面から試みた。

ヘモキュウによるHb測定は，自動血球計算装置との相関で高い相関があり，とくに問題がないことが示された。これは過去の報告のとおりである[8)～10)]。また，比重法とHb法で献血不適格者の比率が異なるか否かを検討した。比重法とHb法の比較検討では，時期が異なるため厳密な比較ではないが，献血不適格者の増減はなく，現行の採血基準で有意の差はないと思われた。男性のVVRは，軽症でHb法の方が少なくヘモグロビン値以外の原因が考えられる。

Hb法の利点は，献血者のHb値を数字として表示できることであり，度数分布を知ることができる。この度数分布によって，女性献血申込者の中に，10g/dL未満の要加療者が不適格者の10%近くみられることが判明した。従来の比重法では，低比重以外の情報がなくそのまま放置されるわけであるが，Hb法ではHb値を提示できるので医療機関への受診を勧めることができた。また，10.0～12.5g/dLの方には栄養指導や食事のアドバイスができた。すなわち，貧血の予防と治療の双方を区別して指導することが可能である。

採血基準では，真性赤血球増加症(多血症)は採血しないことになっているが，比重法ではHb高値者を除外することができない。Hbを測定することによって，17g/dL以上は男性で3.7%にみられ，女性にはみられなかった。また，これらは白血球数，血小板数，赤血球指数が正常で，相対的(ストレス)赤血球増加症と考えられた。真性赤血

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球増加症は白血球増加，血小板増加，小球性低色素性赤血球の傾向を示すことから，今回の検討で，Hb19.0g/dL未満で白血球数，血小板数，赤血球指数が正常であれば，採血可能と判断した．

今回Hb測定の定量性を生かして，従来の採血基準の妥当性を検討した。まず，比重法とHb法の比較で，1.052以上はHb12.1g/dL以上を，1.053以上はHb12.6g/dL以上を示した。また，Hb値の低下に伴って赤血球指数が低下してくるが，平均値の低下開始に相当するHb値は，小球性低色素性赤血球に移行する点で，女性の成分採血の際の可否判定に用いられているところである。低下開始点は男性12.5g/dL，女性12.0g/dLで，男性が0.5g/dL高かった。また，12.5g/dL以下の男性献血申込者の比率は0.6%と少なく，あえて男性の採血基準を引き上げる必要はないと考えられる。以上および米国FDAの基準[11]を勘案して，私たちはHb法の判定に男女差を設けず，従来の採血基準を用いることで問題がないと考えた。

今回用いたヘモキュウによるHb測定法は，英国のNational Quality Assessment Schemeの精度管理で正確性の保証が得られている[11]。また，静脈血採血と耳朶あるいは指尖毛細血管穿刺との間に差異があるとの議論がある。これは，サンプリングが不適切な場合で，血流が十分保たれ，穿刺が正確に行われた場合は有意の差がないとの見解が一般的である[12]。また，指尖穿刺の方が，静脈穿刺より正確性を欠くとの報告もある[13]。

献血の可否を決定する検査は，大別して，血液学的検査，生化学検査，感染症関連検査が行われている。生化学，感染症関連検査は1953年血液事業が開始されて以来，次々と改良，改善が加えられ，NAT検査の導入によって世界的水準を保つにいたっている。一方，採血基準の根幹である貧血の有無判定については，当初の硫酸銅による比重法が現在にいたるも用いられ，一向に改良の気配がない。その間，比重不足による献血不適格者は増加の一途であり，女性の400mL献血で本社の調査で，1990年 9.9%，2000年 18.1%，2003年 21.3%である[14, 15]。輸血によるウイルス性肝炎が激減したのと極めて対照的である。いうまでもなく比重法は測定者の目視による定性的判定法であり，温度・湿度の影響，使用滴下回数や蒸発，観察者の主観を無視できない。臨床の場においても，かつては比重法や比色法(ザーリ法)が用いられたが，現在はHb,ヘマトクリットに統一され，比重，比色によっている医療機関は皆無である。したがって，血液センターと医療機関の間で貧血に関するかぎり整合した議論が全くできていない。国は献血者の確保の推進として，献血の検査結果を健康診査，人間ドック，職場検診で活用するとともに，地域の保健指導に用いるよう求めているが[16]，比重で表示される献血不適格者の成績は利用し得ない状況である。以上から，血液センターにおいてもHb法を早急に導入し，定量的な評価によって献血者の健康を守る配慮をすべきである。

結　論

1．献血の可否判定にHb法を導入した。従来の比重法に比して，不適格者率，副作用発症率とも差異はなかった。
2．Hbおよび赤血球指数の度数分布から，従来の採血基準(400mL：12.5g/dL以上，200mL：12.0mg/dL以上)を用いて差し支えないことが判明した。
3．Hb低値の献血申込者に対して，Hb値に応じた栄養指導，医療機関への受診指導を行うことができた。
4．Hb法は定量性，客観性において比重法に優っており，Hb法に統一すべきであることを提言した。

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(12)

Statistical analysis of inappropriate results from current Hb screening methods for blood donors

Virge James, Keith F. Jones, Elizabeth M. Turner, and Robert J. Sokol

BACKGROUND: The objective was to apply statistical analysis to the false passes and fails that occur with the primary and secondary Hb-screening methods used at blood-donor sessions.

STUDY DESIGN AND METHODS: Venous samples from 1513 potential donors who had undergone primary $CuSO_4$ screening using capillary blood (Hb cut-offs: women, 125 g/L; men, 135 g/L) were tested at the session by a secondary method (HemoCue; cut-offs: women, 120 g/L; men, 130 g/L) and again at the base laboratory using another system (Beckman Coulter General S system), which generated the "true" Hb value.

RESULTS: False-pass and -fail rates for women and men, respectively, were 11.2 and 6.3 percent (women) and 5.2 and 1.8 percent (men) for $CuSO_4$; 1.9 and 3.7 percent (women) and 1.5 and 0.4 percent (men) for HemoCue; and 2.7 and 2.4 percent (women) and 1.8 and 0.2 percent (men) for a combined procedure that mimicked current practice of only testing $CuSO_4$ fails by HemoCue.

CONCLUSION: $CuSO_4$ Hb screening gives large numbers of false passes, particularly in women. Using venous samples, the majority correctly pass at the lower HemoCue cut-offs. The current dual-testing policy appears convenient for donor sessions, but because small percentages of false passes and fails represent large numbers of donors, every effort should be made to improve the accuracy of Hb screening.

Potential blood donors who attend donor sessions in the Trent Region (situated in the East Midlands, UK) initially undergo a health-screening survey. After passed this survey, they are subjected to primary Hb screening by the $CuSO_4$ gravimetric method carried out on finger-prick capillary blood, the cut-off levels for donation being set to correspond to Hb values of 125 g per L for women and 135 g per L for men.[1-3] To optimize blood-collection rates, UK regulations allow individuals who fail the primary $CuSO_4$ test to continue with the donation process if they pass the secondary Hb screening performed on a predonation venous sample using the HemoCue system.[2,4,5] With this method, donor acceptance or rejection is set at lower Hb levels: 120 g per L for women and 130 g per L for men.

We have recently become concerned that some donors are being bled inappropriately with these screening methods, whilst others with an acceptable Hb level are failing the tests. The purpose of this study is to determine whether this is the case and how to quantitate the problem by applying statistical analysis to the primary and secondary Hb-screening procedures used at our donor sessions, comparing them with a standard Hb measurement.

MATERIALS AND METHODS

Studies were carried out on potential volunteer blood donors attending routine donor sessions held throughout the Trent Region. All participants were fully informed of the purpose of the project and gave signed consent. The

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study had been formally approved by the Trent Multicentre Research Ethics Committee.

To avoid bias when selecting individual subjects for the study, a simple systematic sampling scheme was used at each donor session. Before screening, every n^{th} potential donor was approached for consent to enroll in the trial. If an individual declined, each subsequent person was approached until one consented. Subsequently, the next n^{th} individual was approached and so on. The value of n was controlled by the transfusion service staff at the screening station.

During quiet periods, n could be set at 1 so that every potential donor could be approached. During busier periods a larger value of n could be set, and at exceptionally busy times, sampling could be discontinued completely to avoid delaying the session.

Venous blood samples were collected from 730 women and 783 men who were potential donors who had undergone the primary $CuSO_4$ gravimetric Hb-screening test. All the venous samples, which included those from individuals who passed and failed $CuSO_4$ screening, were taken before any blood donation and tested at the donor session by the HemoCue method. These machines are calibrated to the International Council for Standardization in Haematology standard. The HemoCue results were used to construct a hypothetical screening test and were expressed as either a pass or fail in respect to cut-off Hb values of 120 g per L for women and 130 g per L for men.

A combined procedure that followed current practice was also applied. Thus, respondents were initially screened on the standard $CuSO_4$ test; those who passed were deemed to have passed the combined procedure. Those who failed the $CuSO_4$ test were considered to have passed the combined procedure if a subsequent HemoCue result was at least 120 g per L for women and 130 g per L for men.

The venous samples were tested again at the base laboratory with the Beckman Coulter General-S system (Beckman Coulter, High Wycombe, UK). These results were deemed to be the "true" Hb values against which the results of the $CuSO_4$, HemoCue and combined procedures could be compared.

Statistical methodology

In view of the known differences in Hb levels between men and women, data for the different sexes were analyzed separately. Because donor characteristics would be likely to vary considerably between individual donor sessions, any sampling biases with respect to donor age were adjusted by stratifying data for both men and women into quinquennial age bands and then testing to determine whether reweighting of the age-stratified data was necessary. This was achieved by chi-squared tests, comparing test and whole donor population data, and by a one-way ANOVA conducted for each of the women and men data sets with various Hb counts as the dependent variable and age category as the factor of interest.

The need to reweight was confirmed by both tests. A chi-squared value of 54.88 ($p < 0.0001$, df = 10) in respect to age distribution for women indicated that the test sample was severely under-represented in the 17 to 30 years age range, whereas for the age distribution for men, a chi-squared value of 18.60 ($p < 0.046$, df = 10) showed the test sample was under-represented in the 20-and-under ages. For the ANOVA, F values of 3.00 (df = 10,724, $p = 0.001$) for women and 2.23 (df = 10,782, $p = 0.015$) for men confirmed that in each case, Hb varied with age.

Reweighting to give reasonable donor population estimates was therefore carried out by calculating the stratified sample proportion of individuals possessing the appropriate attribute, together with its SE. This proportion is an unbiased estimator of the true population proportion possessing the desired attribute.[6,7] All values and standard errors were obtained using a statistical software package (SAS, SAS Institute, Cary, NC), and all proportions and standard errors were converted to percentages by multiplying them by 100.

The results of each screening test were compared to baseline Beckman Coulter Hb values of 125 g per L (women) and 135 g per L (men) for the $CuSO_4$ test and 120 g per L (women) and 130 g per L (men) for the HemoCue and combined procedures. The "false-pass" rates (i.e., the percentages of potential donors who would pass the relevant screening test but would fail the baseline Beckman Coulter test) were of particular interest.

RESULTS

Table 1 shows the results of the $CuSO_4$ Hb screening compared with the baseline Beckman Coulter values of 125 g per L (women) and 135 g per L (men). Table 2 (women)

TABLE 1. Results of $CuSO_4$ screening test compared with Beckman Coulter baseline at Hb levels of 125 and 135 g per L for women and men, respectively: population percentage estimates, stratum weighted by age

		Women		Men	
$CuSO_4$ result	Beckman Coulter result	Estimated percentage	SE	Estimated percentage	SE
Fail	Fail	12.4	1.3	3.9	0.7
Fail	Pass	6.3	0.9	1.8	0.5
Pass	Fail	11.2	1.3	5.2	0.8
Pass	Pass	70.1	1.8	89.0	1.1
Correct classification (%)		82.5		93.0	

TABLE 2. Results of screening tests for women compared with Beckman Coulter baseline Hb level of 120 g per L: population percentage estimates, stratum weighted by age

Screening test result	Beckman Coulter test result	$CuSO_4$ Estimated percentage	$CuSO_4$ SE	HemoCue Estimated percentage	HemoCue SE	Combined Estimated percentage	Combined SE
Fail	Fail	6.0	1.0	6.0	0.9	5.3	0.9
Fail	Pass	12.7	1.3	3.7	0.7	2.4	0.6
Pass	Fail	1.9	0.6	1.9	0.6	2.7	0.7
Pass	Pass	79.4	1.6	88.4	1.3	89.6	1.2
Correct classification (%)		85.4		94.4		94.9	

TABLE 3. Results of screening tests for men compared with Beckman Coulter baseline Hb level of 130 g per L: population percentage estimates, stratum weighted by age

Screening test result	Beckman Coulter test result	$CuSO_4$ Estimated percentage	$CuSO_4$ SE	HemoCue Estimated percentage	HemoCue SE	Combined Estimated percentage	Combined SE
Fail	Fail	2.2	0.5	2.0	0.5	1.7	0.5
Fail	Pass	3.6	0.6	0.4	0.2	0.2	0.2
Pass	Fail	1.3	0.4	1.5	0.4	1.8	0.5
Pass	Pass	93.0	0.9	96.2	0.7	96.3	0.7
Correct classification (%)		95.3		98.2		98.0	

and Table 3 (men) give the results of the individual $CuSO_4$ and HemoCue screening tests and of the combined procedures, comparing them with Beckman Coulter baseline values of 120 g per L for women and 130 g per L for men.

DISCUSSION

The UK requires a predonation Hb screening to be carried out on all potential donors, and only individuals with an Hb level at or greater than 120 g per L for women or 130 g per L for men proceed to donate.[8,9] However, accuracy of Hb-screening procedures at blood-donor sessions may be a problem, and our study, by quantitating this, provides data for informed debate (Tables 1-3). It also shows how such studies may be approached in the future. In the present case, statistical analysis without the need to reweight would have required an even larger sample size. This would have been impractical because the length of time it took to obtain the informed consent required by the Ethics Committee had a deleterious effect on the efficient running of many donor sessions, particularly busy ones. As a result, the test sample was not representative of the donor population as a whole. This, and because of clustering of sessions, made it important to reweight the data so that the test population truly reflected the whole donor population with regard to factors that affect screening outcomes, such as age and sex. Reweighting necessitated expressing the results in proportions (percentages) rather than as raw figures.

The primary purpose of Hb screening is donor protection, preventing an anemic individual from exacerbating their condition with potential ill effects. The secondary purpose is to ensure the patient receives a minimum infused Hb dose per RBC transfusion. Screening also acts as a nonspecific measure of the general health of the donor and may identify some conditions which could potentially be harmful to the recipient.[2]

Protocols with set cut-offs are not without problems: they cause administration and quality control costs, donor inconvenience, expense and anxiety as a result of medical follow-up of deferrals, as well as permanent loss of donors. Additionally, cut-offs need to be set to maximize donor safety but be balanced against the system's ability to collect an adequate blood supply, a particular concern when trying to exclude women with iron deficiency. Hb reference ranges vary with age, race, and sex, and are affected by altitude, smoking, and the site from which the sample is taken.[2,10] It has been suggested that, rather than having set cut-off values, a standard should be established whereby blood donations contain a "minimum Hb dose" of 50 g; this would allow individual blood centers to evaluate the appropriate safe Hb cut-off for their donors.[2]

The $CuSO_4$ gravimetric test has been the method of choice in the UK for primary Hb screening of potential blood donors for many years. It is fast, inexpensive, does not require a venous sample, and, although rigorous training and constant monitoring of session staff is necessary, does not need trained laboratory personnel. It does not, however, give a quantitative result, has a subjective endpoint, is difficult to quality control, and presents problems with the disposal of biohazardous material.[2] Although very anemic donors can, on occasion, pass the $CuSO_4$ test,[11] early reports suggested that the $CuSO_4$ method tended to give inappropriate failures, and thus significant numbers of such failed donors could be recovered with a revised Hb range or if an alternative screening method was applied.[2]

This is the rationale for the primary and secondary Hb-screening tests used in the UK. It is supported by several studies that show that many units of blood can be collected that would otherwise be lost. Figures of between 11 and approximately 50 percent recovery of donations with secondary screening are quoted.[2,12-14] The lowering of the cut-off Hb values for the secondary screening also helps. In one study, 29 percent of failed

donors passed the secondary test (HemoCue) at Hb cut-offs of 125 and 135 g per L (women and men, respectively); but with the cut-offs reduced to 120 and 130 g per L, this figure increased to over 44 percent.[14]

Initially there was concern that such a high proportion of donors, 11.2 percent of women and 5.2 percent of men in the present study, inappropriately pass the $CuSO_4$ screening test (Table 1); and, it should be noted that at these higher baselines, a HemoCue screening test would have considerably reduced the false-pass rates. Thus, the high false-pass rates in Table 1 do not mean that there is a similar proportion of donors being bled inappropriately. Examination of Tables 2 and 3 show that at baselines of 120 and 130 g per L, the $CuSO_4$ screening tests exhibit conservative false-pass rates similar in magnitude to the HemoCue procedure; only 1.9 percent of women and 1.3 percent of men who pass the $CuSO_4$ test have Hb levels less than 120 and 130 g per L, respectively, and should have been rejected as donors, indicating that, in practice, the current $CuSO_4$ cut-off levels can be tolerated. (The higher false-fail rates with the $CuSO_4$ test in Tables 2 and 3 are due to the higher cut-off settings.)

Tables 2 and 3 show that, had it been used in isolation, the HemoCue procedure would have classified 94.4 percent of women and 98.2 percent of men correctly at Hb levels of 120 and 130 g per L, respectively. Although this would appear to offer an improvement on the $CuSO_4$ test (set at 125 and 135 g/L for women and men, respectively), at present, the HemoCue procedure would be difficult to apply as a primary screening test on every potential donor because venous samples are preferred at our sessions. (HemoCue can be used on finger-prick blood, but capillary samples are known to give unreliable results[12,15] with all technologies and are thus unsuitable for secondary screening of blood donors.) Taking a venous sample from each person before donation could prove unacceptable to donors, slow down the donation process, as well as increase costs. Many studies have shown the excellent correlation between HemoCue and standard photometric methods in the laboratory,[14-18] and indeed we found the same in a prestudy evaluation of the analyzers used in this project. (In addition, HemoCue has a theoretic advantage over other photometric methods in that it incorporates a turbidity control, allowing more accurate results on lipemic samples.[2]) However, previous work has shown that accurate measurement of Hb level using the HemoCue system is difficult to achieve in the field.[19,20] There are several possible reasons for this; they include inadequate mixing of specimens,[19] sampling techniques, and operator performance,[20] rather than problems inherent to the methodology, and studies have shown that meticulous attention to sample mixing, mode of filling the cuvette, and continuous monitoring and training of staff can help to improve performance.[20]

Tables 1 through 3 show that the $CuSO_4$ and HemoCue screening tests are less accurate, compared with Beckman Coulter values, for women than men, with false-pass and -fail rates being higher for women than males. This has been recognized previously, and it was suggested that such differences in screening-test performance can be explained by the distribution of women and men donor Hb levels relative to the cut-off values for acceptance.[21] A comforting factor in our study, in spite of its relatively small sample size, is that the lowest false-pass levels were 109 g per L for women and 123 g per L for men. Although it was inappropriate to collect blood from such individuals by our current guidelines, these figures are not alarming; there were no clinical sequelae, as far as we are aware, in the donors, and the recipients would have obtained an adequate amount of Hb. The donors who had been inappropriately bled were contacted and informed.

The results of the "combined" screening procedures (Tables 2 and 3), which mimic current practice at donor sessions, respectively, show false-pass and false-fail rates of 2.7 and 2.4 percent, respectively, for women and 1.8 and 0.2 percent, respectively, for men. The false-pass rates for the combined procedure slightly exceed those for the HemoCue alone: 95-percent CIs for these differences in rate are approximately 1.6 and 0.8 percent for women and men, respectively. On the other hand, the false-fail rates on the combined procedures are slightly smaller than for HemoCue alone, with 95-percent CIs for these differences in rate of approximately 2.3 and 0.6 percent for women and men, respectively. It should be noted here that any false pass on HemoCue alone would also pass the combined procedure, regardless of the $CuSO_4$ test result. Consequently, the false-pass rate for the combined procedure must be at least as great as that for HemoCue alone.

In summary, compared with HemoCue alone, current practice trades off a slightly higher false-pass rate against a slightly lower false-fail rate, and so is still reasonable in spite of the error rates in the initial $CuSO_4$ screen, and they need not be changed until the problems of accurately measuring Hb in the field can be reduced or eliminated. Because approximately 2 million donations are collected annually in the UK, even small percentages of false passes and false fails at the Hb-screening stage represent a large number of individuals, and, consequently, any improvement in accuracy of Hb screening will be welcome.

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13

原 著

短期間の術前自己血貯血法の検討

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はじめに

医療レベルの向上に伴いその質が問われる現在，手術における同種血輸血の回避は患者の当然の選択肢となりつつある。心大血管手術においては，早くから多くの施設が積極的に自己血輸血を導入することにより，無輸血達成へ向けて努力している。無輸血達成率は自己血貯血量および貯血期間に比例するのは周知の事実である。しかし心大血管手術においては，長期の待期期間を設けられる場合がそれほどなく，術前の長期入院や通院も患者の負担が大きい。そこで当科では可及的に貯血期間を短縮し，貯血量を最大限に準備できる方法として，術前8日からの貯血開始を基本的に施行してきた。今回この貯血法を施行した186例を貯血期間が9日以上であった群と7日以下であった群とで比較検証し，また同種血輸血に至った例と無輸血例とを要因別に比較し，その成績と限界について検討した。

対象・方法

当科で1996年9月から2003年2月までに人工心肺を使用した心大血管手術例は427例であった。そのうち自己血貯血を施行したのは258例で，すべての人工心肺使用例中60.4％，全待期手術中73.5％あった。対象手術は冠動脈バイパス術，弁膜症手術，胸部大動脈瘤手術，先天性心疾患手術，その他であった。自己血貯血の適応は，原則として年齢が80歳以下で入院時Hbが10.0 g/dl以上の待期手術としており，非適応は感染性心内膜炎患者，透析患者，高度心不全患者，左主幹部病変を伴う不安定狭心症患者としている。貯血は全例入院中としている。自己血採血のプロトコールは，毎回採血前にHb値を測定し，10.0 g/dl以上であれば1週間ごとに400 ml採血している。保険適応内であればエリスロポエチン製剤（EPO）を6,000単位静脈投与を隔日投与，もしくは24,000単位の皮下注投与を隔週に投与した。また，鉄剤としてフマル酸第一鉄305 mgを毎日内服投与した。ここで論ずる貯血期間とは，初回自己血貯血開始日より手術前日までの日数とした。待期手術の患者は8日前に入院し，入院日に400 mlの貯血を行い，1週間後の手術前日にも400 ml貯血する（EPO投与は皮下注の場合は初回の1回のみ，静注の場合は計3回投与となる）という貯血法を186例に施行した（M群）。準緊急手術症例や心房中隔欠損症などの軽症例では貯血期間が7日以下で，400 mlのみの貯血で手術に臨み，これらは44例であった（S群）。術前の精査などで術前8日以前より入院可能であった患者においては，手術が決定した時点から貯血を開始した。このような症例で9日以上の貯血期間が得られたのは28例であった（L群）。これらの3群の無輸血率を比較するとともにM群において同種血輸血に至った例と無輸血例を性差，年齢，体重，EPO使用量，入院時Hb値，手術直前Hb値，人工心肺時間，手術時間，術式についておのおの要因別に比較した。検討において，

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術後から退院まで同種血輸血を施行しなかったものを無輸血例とした。手術時は全例回収洗浄式自己血輸血装置を用い，術後約12時間はドレーン排液も回収した。人工心肺は無血体外循環で手術終了時回路内血液を返血した。各群の数値は平均値±標準偏差で表し，統計学的検定はstudent-*t*, χ^2, 分散分析を用い，p値＜0.05を有意差ありとした。

結果

各群の手術術式の内訳，およびその無輸血率は表1に示した。冠動脈バイパス術に貯血期間が短い傾向がみられたが，手術を急ぐ必要のある例が多かったためと思われた。おのおの3群間に有意差は認めなかったが，冠動脈バイパス術の無輸血率が低く，貯血期間の短い群にその傾向が強かった。各群の性差，年齢，体重，貯血期間，総貯血量，EPO使用量，入院時Hb値，手術直前Hb値，人工心肺時間，手術時間，無輸血率を表2，表3に示した。S群の貯血期間は1～7日，平均5.5±1.6日で，L群が9～28日，平均15.8±5.6日であった。総貯血量はM群で400～800 ml，平均770±103 ml，S群がすべて400 ml，L群が800～1,600 ml，平均1,029±249 mlであった。性差，

表1　対象手術と無輸血率

術式	例数			無輸血率		
	M群	S群	L群	M群	S群	L群
CABG	72 (63.7%)	29 (25.7%)	12 (10.6%)	72.2%	55.2%	91.7%
VD	76 (78.4%)	8 (8.2%)	13 (13.4%)	90.8%	87.5%	92.3%
TAA	14 (87.5%)		2 (12.5%)	78.6%		100 %
CHD	12 (63.2%)	7 (36.8%)		100 %	100 %	
その他	12 (92.3%)		1 (7.7%)	66.7%		100 %

CABG:冠動脈バイパス術，VD:弁膜症手術，TAA:胸部大動脈瘤手術
CHD:先天性心疾患手術

表2　対象群の比較1

	例数	性差 (M/F)	年齢 (years)	体重 (Kg)	貯血期間 (days)	総貯血量 (ml)	EPO投与量 (×1000 IU)
M群	186	119/67	63.1 ± 12.9	56.3 ± 9.1	8.0 ±0.0	770 ± 103	20.9 ± 5.9
S群	44	28/16	62.7 ± 10.4	57.3 ±10.9	5.5 ±1.6	400 ± 0	3.8 ± 7.4
L群	28	18/10	61.6 ± 9.1	59.6 ± 9.6	15.8 ±5.6	1029 ± 249	29.4 ± 15.3

表3　対象群の比較2

	入院時Hb (g/dl)	手術直前Hb (g/dl)	人工心肺時間 (min.)	手術時間 (min.)	無輸血率	p value
M群	13.0 ± 1.4	11.0 ± 1.4	114 ± 70	246 ± 124	81.7%	M群 vs S群: 0.047*
S群	12.9 ± 1.7	11.4 ± 1.4	99 ± 49	242 ± 155	68.2%	
L群	13.5 ± 1.3	11.2 ± 1.4	109 ± 35	223 ± 53	92.9%	M群 vs L群: 0.231

年齢，体重，入院時Hb値，手術直前Hb値，人工心肺時間，手術時間において3群間に有意差は認めなかった。M群の無輸血率は81.7%で，S群の68.2%と比べ有意に高く（p=0.047），L群の92.9%と比べ低いものの有意差はなかった。M群において同種血輸血例と無輸血例を，性差，年齢，体重，貯血量，EPO使用量，入院時Hb値，手術直前Hb値，人工心肺時間，手術時間の各要因で比較したところ（表4），年齢，体重，入院時Hb値，手術直前Hb値，人工心肺時間，手術時間において有意差を認めた。M群の内で，2回目の採血前にHb値が10.0 g/dl以下，もしくは全身状態不良，採取困難な例で800 ml貯血できなかった例は15例（8.1%）あり，その無輸血率は66.7%と低い傾向にあったが，800 ml貯血例の無輸血率と有意差は認めなかった。また，術後出血再開胸や再手術を施行した例は9例あり，その無輸血率は44.4%と有意に低かった。術式では冠動脈バイパス術と弁膜症手術を比較すると前者で無輸血率が有意に低値であった（表5）。なお，全例において自己血廃棄例はなかった。

考察

心臓血管外科領域においては，他の領域に先がけて早くより同種血輸血回避に対する努力が試みられ，年々手術成績が向上するに伴い無輸血手術に対する関心は広がりつつある。無輸血達成へのもっとも効果的な方法として，術前貯血式自己血輸血が施行されるようになり[1]，人工心肺を使用する心大血管手術においては，現在ほぼ一般的な手法とされている[2]。しかしその適応や貯血期間

表4 M群における輸血例と無輸血例の要因別比較

要因		輸血例	無輸血例	P値
男女比	(M/F)	17/17	102/50	0.060
年齢	(years)	69.4 ± 8.2	61.7 ± 13.3	0.002 *
体重	(Kg)	51.7 ± 8.5	57.3 ± 9	0.001 *
貯血量	(ml)	741 ± 144	777 ± 91	0.067
EPO使用量	(×1000IU)	21.9 ± 4.6	20.6 ± 6.1	0.269
入院時Hb	(g/dl)	12.5 ± 1.5	13.1 ± 1.4	0.032 *
手術直前Hb	(g/dl)	10.0 ± 1.1	11.2 ± 1.4	<0.001 *
人工心肺時間	(min.)	173 ± 123	101 ± 42	<0.001 *
手術時間	(min.)	381 ± 211	216 ± 64	<0.001 *

表5 M群における無輸血率に影響する因子

	例数	輸血例	無輸血率	p value
800ml未完遂	15 (8.1%)	5	66.7%	
800ml完遂	171 (91.9%)	29	83.0%	0.221
再開胸(+)	9 (4.8%)	5	44.4%	
再開胸(-)	177 (95.2%)	29	83.6%	0.012 *
冠動脈バイパス術	72 (38.7%)	20	72.2%	
弁疾患手術	76 (40.9%)	7	90.8%	0.003 *

に関しては，施設間で一定していないのが現状である。施設間で手術方法，成績，麻酔科の方針，病院での輸血に対する取り組み，マンパワー等，あらゆる面で異なるので，自己血貯血に対する方針にも若干差が見られて当然である。長期の貯血期間を設け，多量の貯血量を準備できれば，無輸血率が飛躍的に向上するのは当然のことである。しかし心大血管手術においては，それほど長期の待期期間を経て手術となる症例は少ない。また病院の稼働率を考慮した場合，術前の入院期間は制約を受けるのが現状である。外来通院での貯血は理想的であるが，輸血部のようなユニットが独立している大規模な施設以外では，マンパワーの制限があったり，心疾患患者での外来採血は不安も多く，患者の術前の精神的負担も大きい。したがって，当施設もそうであるが，入院後の自己血貯血が原則となる。自己血貯血にEPO投与が効果的であることは多く報告され[3,4]，ほとんどの施設で使用されているが，保険基準で貯血量が800 ml以上で1週間以上の貯血期間が必要と定められている。この基準を満たし，かつ最短の貯血期間を設けるため，当科では術前8日からの入院および貯血開始を施行してきた。無輸血率は81.7％とある程度許容される成績ではあるが，やはり貯血期間の長い症例と比較すると，有意差はないものの低い傾向にあった。しかし貯血期間が1週間以内で，400 mlしか貯血できなかった症例（S群）よりは有意に良好な無輸血率であった。開心術にあえて貯血式自己血輸血をせず，良好な結果を示した報告もある[5]。しかし同種血輸血の安全性が100％確立されていない現在，多少とも自己血貯血やEPO投与の機会があり，無輸血の可能性が1％でも増えるならば，その選択肢は提供されるべきであろう[6]。この貯血法で同種血輸血に至った症例は，無輸血例に比べ，高齢で低体重，術前のHb値が低いという結果は当然考えられ，人工心肺時間および手術時間の長い例ほど輸血率が高いという結果も他の報告と同様であった[7]。この短期間で800 mlの採血は手術直前のHb値が他の報告に比べ著しく低く，平均が11.0 ± 1.4 g/dlであった。つまりEPO投与で，十分な造血効果が発揮されるには期間が短すぎるかもしれない。エリスロポエチンによる造血刺激を促すには最低3週間必要という報告も見られる[8]。しかし，われわれは以前1週間でも造血効果は有意に上がっている結果を報告している[4]。初回の開心術における貯血量は800 mlが至適であるという報告も見られる[9]が，その800 mlを採血した後の手術直前Hb値がどれだけ保たれているかも重要な要因と思われる。これは貯血期間と造血能に依存し，この術直前Hb値の低さはこの貯血法の限界であろうと考える。しかし術前の患者の全身状態に影響がない限り，手術前日でも400 mlの貯血は無輸血手術に有効と考える。出血再開胸や他の再手術を要した症例の無輸血率は著しく低かったが，これらの症例は貯血期間，量に関係なく同種血輸血を要したと考えられるので，初回手術に限れば無輸血率はもう少し良好と思われた。また，冠動脈バイパス術の無輸血率が弁膜症手術に比べ有意に不良であったのは，前者の方がバイパスグラフト採取などで有意に手術時間が長いこと，術前に抗凝固剤が投与されている例も多く，出血量が多いためと考えられた。この貯血法の妥当性を検討した場合，単独弁膜症手術，心房中隔欠損閉鎖術など，比較的人工心肺時間や手術時間の短い症例であれば，ほぼ満足すべき結果が得られる方法と思われた。少量の貯血量で十分と予想されても予想外に侵襲，出血が多くなることもあり，無輸血手術を第一義的に考えれば「最大限の貯血期間を設け，できうる限り多量の貯血を行う」ということに尽きると思われる。しかし，同種血無輸血を目指すあまり，患者に術前の負担を過剰にかけたくないという方針で，当科ではこのような貯血法を基本とした。すべての開心術に有効とはいえないまでも，長期の待期期間が設けられない症例に対し，比較的短期間の術前入院および貯血期間でほぼ良好な無輸血率を達成できる一手法として，今後も活用したいと考える。

結語

人工心肺を用いる心大血管手術において，貯血期間8日で800 mlを貯血する自己血貯血法を186例に施行し，無輸血率81.7%と比較的良好な成績を得られた。

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原 著

自己血400 ml採血後2週間のヘモグロビン値の回復度に与える影響因子の検討

眞鍋 庸三・瀬戸 美夏・冨永 晋二・谷口 省吾

はじめに

我々は顎矯正手術の一つである上下顎同時移動術に対して，400 ml採血の貯血式自己血輸血と希釈式自己血輸血を併用し，同種血輸血を100 %回避している。本手術の対象となる患者は若くて健康であるが，低体重の女性が多いという特徴がある。当院では，自己血採血によって低下するヘモグロビン値（Hb値）をはじめとする種々の因子の回復を考慮し，予定手術日の3週間前に採血することを原則にしている[1]。しかし，患者の都合などにより術前2週間の採血を余儀なくされる症例もある。今回，手術2週間前に採血を行った患者のヘモグロビン値の回復度に影響を与える因子について検討した。

対象・方法

福岡歯科大学付属病院において，文書と口頭にて自己血輸血の説明を行い同意が得られ，手術の約2週間前に400 mlの自己血採血を施行した患者47名（男性13名，女性34名）を対象とした。自己血採血当日の採血前に検査血を採取し，Hb値，血清鉄値，フェリチン値，総鉄結合能（TIBC），不飽和鉄結合能（UIBC），血清総蛋白量（TP），血小板数（Plt），白血球数（WBC）を測定した。400 mlの自己血採血を行った後，フェジン® 80 mgを加えた1000 mlの晶質液輸液または300 mlの膠質液輸液を行い，さらに採血翌日から2週間，200 mg/dayの鉄剤を経口投与した[2]。入院後，手術前日に検査採血を行い，この時のHb値を術直前Hb値とした。循環血液量は，体格や性別によって大きく異なる[3]が，当院では，一律400 mlの採血を行っているため，採血後のHb値低下の程度も異なると考えられる。そこで，400 mlの自己血採血後，1000 mlの晶質液輸液または300 mlの膠質液輸液を行った時のHb値を予測できる，当科で用いている計算方法[4]を用いて採血後予測Hb値を算出した。循環血液量（CBV）をOgawa式[5]にて求め，図1に示す式にて採血後予測Hb値および予測Hb値の採血前Hb値に対する割合（α）を算出した。また，実測した術直前Hb値の採血前Hb値に対する割合（β）を求めた。さらに採血前の各検査データとβとの間の相関関係の有無を検討した。統計処理には分散分析（多重比較検定：Scheffe法），χ^2検定およびピアソンの相関係数の検定を用い，危険率1 %未満を有意差有りとした。

結果

対象患者全員の予測Hb値を算出したところ，その平均値は12.4 g/dlであった。採血前の平均Hb

福岡歯科大学診断・全身管理学講座麻酔管理学分野

$$\text{・循環血液量(CBV)}\ (\ell) = \begin{cases} \text{男性}: 0.168\times(\text{身長}_{(m)})^3+0.05\times\text{体重}_{(kg)}+0.444 \\ \text{女性}: 0.25\times(\text{身長}_{(m)})^3+0.063\times\text{体重}_{(kg)}-0.662 \end{cases}$$

$$\text{・採血後予測Hb値}\ (g/dl) = \text{採血前Hb値}\ (g/dl) \times \frac{(CBV-0.4)}{CBV}$$

$$\cdot\ \alpha = \frac{\text{採血後予測Hb値}\ (g/dl)}{\text{採血前Hb値}\ (g/dl)}$$

$$\cdot\ \beta = \frac{\text{術直前Hb値}\ (g/dl)}{\text{採血前Hb値}\ (g/dl)}$$

図1　αおよびβの算出方法
α：採血後予測 Hb 値の採血前 Hb 値に対する割合
β：実測した術直前 Hb 値の採血前 Hb 値に対する割合

表1　A群およびB群の患者背景と採血前検査値

		A群 (n=22)	B群 (n=25)
男女比	(男:女)	5 : 17	8 : 17
年齢	(歳)	23.1±6.9	24.2±3.9
身長	(cm)	162.1±7.9	163.8±11.1
体重	(kg)	53.9±7.3	57.7±13.3
血清鉄	(μg/dl)	88.1±31.1	84.8±27.3
フェリチン	(ng/ml)	51.6±37.5	49.9±57.6
総鉄結合能(TIBC)	(μg/dl)	281.5±38.8	288.4±31.1
不飽和鉄結合能(UIBC)	(μg/dl)	193.4±53.9	205.0±40.7
血漿総蛋白量(TP)	(g/dl)	7.1±0.4	7.1±0.4
血小板数	($\times10^4/\mu l$)	21.4±5.6	23.4±5.3
白血球数	($\times10^2/\mu l$)	54.4±11.2	62.2±14.2
採血前Hb値	(g/dl)	13.3±1.2 *	14.3±1.3

* $p<0.01$　(Mean±SD)

A群：αが0.035以上増加した患者
B群：αの増加が0.035未満であった患者

値は13.8 g/dlであったので，αの平均は0.894となる。また，実測の術直前Hb値は12.8 g/dlであったのでβの平均は0.929となり，対象全員の（$\beta-\alpha$）は，採血後2週間で平均0.035上昇していたことになる。このことからαが0.035以上増加した患者をA群とし，0.035未満であった患者をB群として比較検討した。A群は22名，B群は25名であり，群間の男女比，年齢，身長，体重には有意差は認められなかった。両群間の血清鉄値，フェリチン値，TIBC，UIBC，TP，Plt，WBCに

表2　C群およびD群の患者背景と採血前検査値

		C群 (n=5)		D群 (n=42)
男女比	(男:女)	2 : 3		11 : 31
年齢	(歳)	24.2±11.4		23.6±4.6
身長	(cm)	164.6±8.0		162.8±9.9
体重	(kg)	56.6±4.4		55.8±11.5
血清鉄	(μg/dl)	106.0±46.2	*	84.0±26.0
フェリチン	(ng/ml)	65.2±41.4		48.9±49.9
総鉄結合能(TIBC)	(μg/dl)	278.8±45.6		285.9±33.8
不飽和鉄結合能(UIBC)	(μg/dl)	172.8±86.3	*	202.7±41.0
血漿総蛋白量(TP)	(g/dl)	7.0±0.4		7.1±0.4
血小板数	($\times 10^4$/μl)	20.2±8.1		22.8±5.1
白血球数	($\times 10^2$/μl)	49.8±11.0		59.5±13.4
採血前Hb値	(g/dl)	13.2±1.8		13.9±1.3

* $p<0.05$　(Mean±SD)

C群：βが1以上であった患者
D群：βが1未満であった患者

有意差は認められなかったが，採血前Hb値はA群で有意に低い値を示した（表1）。しかし，採血前Hb値とβの相関関係については，決定係数（0.069），相関係数（－0.093）と共に低く，相関関係は認められなかった。

次に，採血後2週間の術直前Hb値が採血前Hb値以上に増加したC群（β≧1）とそれ以下にしか回復しなかったD群（β＜1）に分配し，比較検討した。C群は5名，D群は42名であった。両群間の患者背景に有意差は認められなかったが，血清鉄およびUIBCに有意差を認めた（表2）。しかし，採血前Hb値を含むその他の採血前検査値に差は認められなかった。血清鉄およびUIBCとβとの相関を見ると，相関係数はそれぞれ0.356，－0.359と低く，両者の間には，弱い相関関係しか認められなかった（表3）。

表3　患者背景および採血前検査値とβとの相関関係

	相関係数	p値
年齢	-0.199	0.226
身長	0.235	0.150
体重	0.135	0.414
血清鉄	0.356	0.026
フェリチン	0.227	0.166
総鉄結合能(TIBC)	-0.207	0.208
不飽和鉄結合能(UIBC)	-0.359	0.024
血漿総蛋白量(TP)	-0.047	0.780
血小板数	-0.096	0.564
白血球数	-0.301	0.062
採血前Hb値	-0.093	0.574

考察

上下顎同時移動術時には輸血が必要となるような出血が起こる場合があり，患者のQOLを考慮すると有効かつ安全な自己血輸血が望まれる。当院における本術式の出血量は，大部分の症例で600～800 mlであるが，1,000 ml以上出血する症例もあるため[1]，確実に同種血輸血を回避するために

は自己血貯血は必須である。我々は，本法に対して400 mlの自己血貯血を行っているが，他施設においても術前の貯血量は400 mlが主流となっている[6]。800 ml以上の貯血を行わないとエリスロポエチンは健康保険の適応外となるため使用できず，採血による貧血を回復させるためには，十分な期間をとる必要性がある。顎矯正外科手術は待機手術であり，大部分の患者は若く，健康状態は良好であるため外来採血が可能で，通常は比較的長く術前貯血期間をとることができるが，患者の時間的な都合や手術日の決定が遅延することなどにより期間を短縮せざるをえない場合もある。他領域の手術においては術前貯血量が800 ml以上必要となるような症例ではエリスロポエチンを併用して手術1週間前まで採血を行い，Hb値の低下もほとんど認められなかったという報告がある[7]。一方で，多少の貧血があっても術前400 ml貯血をした胃全摘術において100 %術中の同種血輸血が回避できたという報告[8]もあり，上下顎同時移動術を受ける患者では400 mlの貯血と術前貯血期間を十分とることで同種血回避率100 %をより確実に維持できると考えられる。

幹細胞の分化が始まって末梢血中に網状球として出現するのに要する期間は，約8日であり[9]，健康成人の生理的赤血球産生量は，全血量に換算すると1日30～40 mlである[10]。さらに，有効な造血刺激が加わると赤血球産生予備能は最大5～6倍まで亢進する[11]。これらのことからは，2週間の貯血期間は貧血回復には十分な期間であるように考えられる。しかし，今回検討した47例中400 ml採血後2週間で完全に元のHb値に回復したものは5例のみであったことから，臨床的には，採血から手術までの期間が2週間以上あることが望ましいと考えられる。症例数が少なかったこともあり，予測因子を明確にすることはできなかったが，採血前のHb値の低い症例の方がβが高かったことから，採血前Hb値が低いほど赤血球造血能が亢進する可能性が示唆された。これは，鉄欠乏性貧血患者は，貯血開始1～2週の早期から著明な造血能の亢進がみられるという新名主らの報告[12]と一致する。この理由として貧血患者では，貧血のない患者と比較して採血後の内因性エリスロポエチン濃度が高い[11]ことが考えられる。しかし，造血には，エリスロポエチンとともに材料となる鉄が必要である。C群の5症例は，D群の42症例と比較して，採血前Hb値には差がなく，血清鉄およびUIBCに有意差を認めた。フェリチン値には有意差は認められなかったが，その平均値はD群が48.9 ng/mlであったのに対しC群では65.2 ng/mlと高い傾向にあった。これは，貧血患者の方が早期のHb値の回復は速いが，完全に回復するためには貯蔵鉄量が関係する可能性がある。採血後全症例に量的には十分な鉄剤を投与しているので，採血前の貯蔵鉄量が関係する可能性は少ないように思われるが，鉄の吸収には個人差があり，採血前に貯蔵鉄量の多い患者の方が鉄の吸収度が高かったことが要因の一つとして考えられる。また，C群の採血前Hb値はD群のそれと有意差がなく，貯蔵鉄量は多かったことから考えて鉄を有効にHb生成に利用できている可能性が考えられる。しかし，今回は鉄剤投与後の貯蔵鉄量を測定していないため明らかにはできなかった。さらに検討を進めることで，顎矯正外科手術に対するより有効な術前貯血を行うことが可能となるものと考える。

結語

術前2週間に400 mlの採血を行った症例においてHb値回復に影響をおよぼす因子について検索した。採血前Hb値と貯蔵鉄量がHb値回復程度に影響を与えている可能性が示唆された。

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15

Daily doses of 20 mg of elemental iron compensate for iron loss in regular blood donors: a randomized, double-blind, placebo-controlled study

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BACKGROUND: A considerable number of regular blood donors develops an iron deficiency, and the exact amount of iron required to compensate for the iron loss from whole-blood donation in males and females is still unknown.
STUDY DESIGN AND METHODS: A total of 526 regular blood donors (289 male and 237 female) were randomly assigned to treatment with either 40 mg, 20 mg, or 0 mg per day of elemental iron as ferrous gluconate for a period of 6 months, during which one unit of whole blood was collected on four occasions (males) or three occasions (females). Hemoglobin level, serum ferritin, and soluble transferrin receptor levels were measured before each donation.
RESULTS: Daily doses of either 40 mg or 20 mg of elemental iron adequately compensated for iron loss in males, who gave blood at 2-month intervals, but did not result in a positive iron balance or an increase in storage iron as reflected by the logarithm of the ratio of transferrin receptor to ferritin concentration. In females, who donated at 3-month intervals, the same daily doses not only restored the iron balance but also led to an increase in storage iron. The number of gastrointestinal side effects due to iron supplementation (12%) was only slightly higher in both iron groups than in the placebo group.
CONCLUSION: The results of this study indicate that 20 mg of elemental iron per day can adequately compensate for iron loss in males and females who donate whole blood up to four (females) or six times per year (males).

The major side effect of whole-blood donation is iron depletion. In Germany, men are generally allowed to donate whole blood every 8 weeks and women every 12 weeks. However, the normal diet is usually unable to compensate for the resulting iron loss.[1,2] Consequently, a considerable number of regular blood donors develops a negative iron balance that may eventually progress to iron deficiency anemia.[3-7] Menstruating female donors are at a particularly high risk for chronic iron deficiency. Although this is well-known, only a few controlled, double-blind studies have dealt with the question of whether iron supplementation can prevent iron depletion in menstruating female blood donors.[8-11] There is evidence suggesting that daily doses of 40 mg of elemental iron as ferrous sulfate can sufficiently compensate for iron loss resulting from whole-blood donation and can improve iron status.[10,11] However, the question of whether a lower dose of iron is sufficient to compensate for iron loss in female donors is still open. In addition, controlled studies on iron supplementation in male donors are lacking. Most importantly, no valid measure of iron storage was used in early studies.[12,13] Today, serum ferritin and soluble transferrin receptor levels can be routinely measured and iron status can be much better assessed than previously.[14-17] The logarithm of the ratio of

ABBREVIATIONS: Fe^{2+} = elemental iron as ferrous gluconate; log(TfR/F) = logarithm of ratio of the soluble transferrin receptor to ferritin concentration.

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the soluble transferrin receptor to ferritin concentration (log[TfR/F]), which was shown to have a highly linear correlation to body storage iron, is currently the most precise measure of body storage iron available.[14,15] Here, we present the results of a double-blind study in which we randomly assigned regular male and female blood donors to treatment with 40 mg, 20 mg, or 0 mg (placebo) per day of elemental iron for 6 months.

MATERIALS AND METHODS

Selection of donors and study design

A total of 526 regular blood donors (289 male and 237 female) were enrolled in this study, which was approved by the Ethics Committee of Charité University Medical Center. Written informed consent was obtained from all volunteers. In accordance with the German guidelines for blood donor selection, all donors were determined to be healthy based on their history and had hemoglobin (Hb) concentrations of no less than 13.5 g per dL (males) or 12.5 g per dL (females). The investigational products consisted of identical capsules in blister packs containing 1.5 mg pyridoxal-phosphate, 2.25 μg cyanocobalamine, 400 mg ascorbic acid, 200 μg folic acid, and 75 μg biotin without (placebo) or with 20 mg of elemental iron as ferrous gluconate (Fe^{2+}) (Phyt-Immun GmbH, Homburg, Germany). Ascorbic acid was added to enhance iron absorption. Because most people believe in beneficial effects of vitamin supplements, the other selected vitamins were added for improved compliance. The form of iron used meets the European Community criteria for dietary foods for special medical purposes. The participants were randomized to one of three groups receiving either 40 mg Fe^{2+}, 20 mg Fe^{2+}, or 0 mg Fe^{2+} in two capsules once daily for 6 months. Hb, serum ferritin, and soluble transferrin receptor levels were determined before blood collection at each initial and follow-up visit. Each male volunteer was scheduled for a total of four visits, including a randomization visit before the first donation at Week 0 and three subsequent predonation visits at 2-month intervals. The females were scheduled for a total of three visits: a randomization visit at Week 0 and two predonation visits at 3-month intervals (Fig. 1). The intervals were chosen in accordance to the German guidelines, which allow six donations per year for male and four donation per year for female volunteers. Volunteers with hemoglobin concentration less than 13.5 g per dL (males) or 12.5 g per dL (females) were deferred, but not excluded from study. Compliance, which was defined as the ingestion of at least 90 percent of the capsules as prescribed, was checked by counting the returned capsules between blood donations.

Laboratory methods

Hemoglobin concentrations in fingerstick blood samples were determined by the acid methemoglobin method using a photometer (HemoCue B-Hemoglobin photometer, HemoCue, Großostheim, Germany). Ferritin and soluble transferrin receptor concentrations in serum were

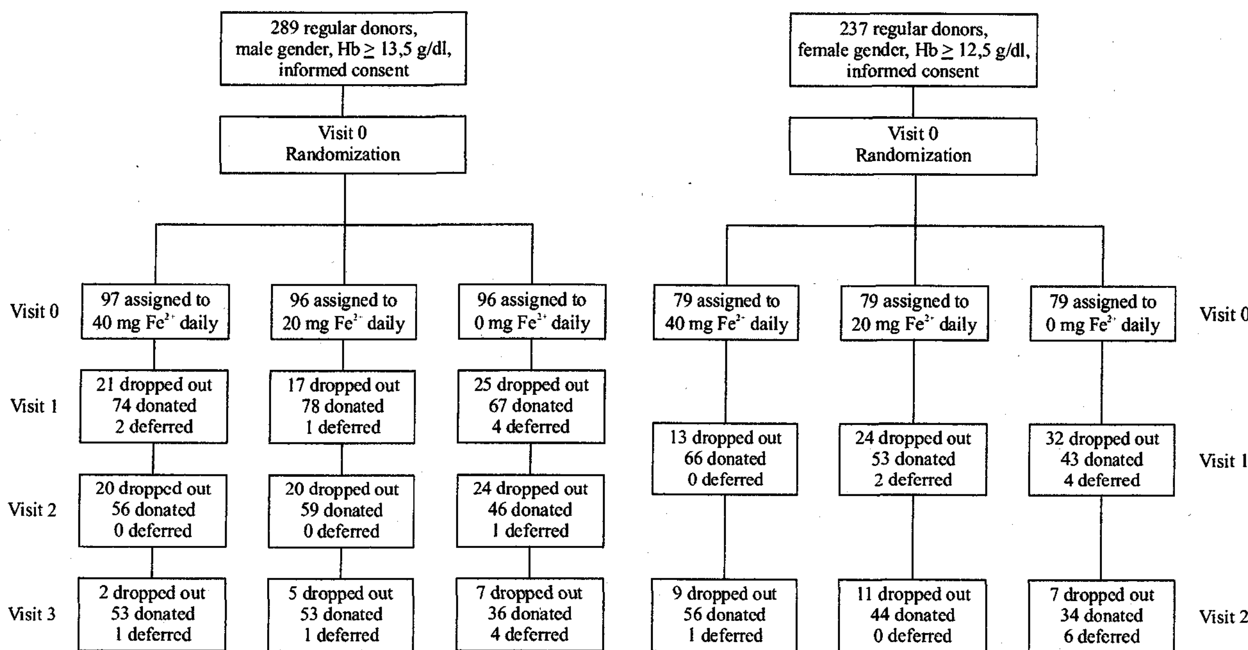

Fig. 1. Flow of participants during study.

determined by nephelometry using an automatic analyzer (BN Prospec, Dade Behring, Marburg, Germany).

Statistics

Sample-size calculation, randomization, and statistical analyses were performed using software (Stata for Windows, Stata Corp., College Station, TX). Based on the serum ferritin concentration, the required sample size was determined to be 49 males and 40 females per group, assuming a power of 0.9, a significance level of 0.0167 (Bonferroni adjustment for three groups), a smallest meaningful ferritin difference of 10 µg per L between groups, three (males) or two (females) follow-up measurements, a within-subject correlation coefficient of 0.8, and a standard deviation (SD) of 26 µg per L (males) or 22 µg per L (female) for serum ferritin. Assuming a dropout rate of 50 percent, we arrived at a final sample size of 98 males and 80 females per group.

The randomization plan was generated using block randomization with variable block length. Statistical analyses were performed as an intent-to-treat analysis for all participants coming for more than one visit using a linear regression model for longitudinal data (cross-sectional time-series regression model with generalized estimating equation analysis).[18] The logarithm of the ratio of transferrin receptor to ferritin concentration, an accepted measure of storage iron, was used as the outcome variable. To model the change in storage iron over time, we applied the difference values for log(TfR/F) and included the iron supplement as the predictor variable.

RESULTS

Males

Of the 289 male volunteers (age range, 19-67 years) enrolled in the study, 141 (49%) dropped out, yielding a dropout rate of 44 percent in the 40 mg of Fe^{2+} group, 44 percent in the 20 mg of Fe^{2+} group, and 58 percent in the placebo group (p = 0.075; Fisher's exact test). A total of 63 (45%) of the male dropouts withdrew before their second visit (Table 1). The mean interval between visits was 60 days. Deferral from donation because of unacceptable hemoglobin concentration values (<13.5 mg/dL) occurred in 14 of 825 visits (1.7%). This was more frequently the case in the placebo group than in the 20 mg and 40 mg iron groups (n = 9 vs. 2 vs. 3, p = 0.022; Fisher's exact test). Compliance was poor in roughly one-third of the male participants.

In the male placebo group, the mean serum ferritin concentration decreased from 35 µg per L at baseline to 21 µg per L at the final visit, the number of males with depleted iron stores (ferritin <12 µg/L) increased from 20 percent to 54 percent, and the mean concentration of soluble transferrin receptors rose slightly from 1.6 mg per L to 1.7 mg per L (Table 2, Fig. 2). In the male 20 mg iron group, serum ferritin decreased from 35 µg per L to 25 µg per L, whereas the median ferritin value changed only slightly (Table 2, Fig. 2); both the number of males with depleted iron stores (25%) and the transferrin receptor concentration (1.5 mg/L) remained nearly constant. In the male 40 mg iron group, the ferritin (33 µg/L) and transferrin receptor levels (1.5 mg/L) remained constant, whereas the number of individuals with iron depletion dropped from 26 percent to 13 percent.

The log(TfR/F) remained nearly constant in both iron groups, but rose continuously in the placebo group

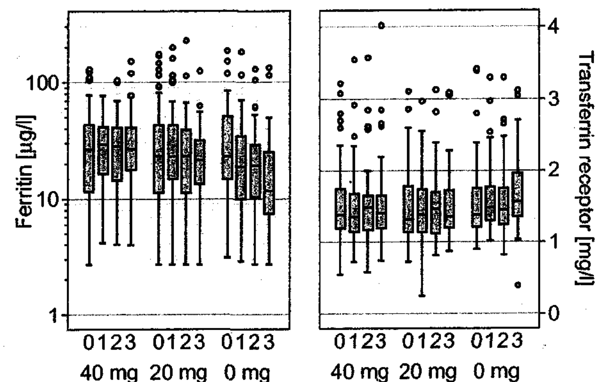

Fig. 2. Box-plot for the concentration of serum ferritin and soluble transferrin receptor in male donors.

TABLE 1. Reasons and numbers of dropouts during study

Reason	Unknown (%)	Unknown (n/total)	Gastrointestinal complaints (%)	Gastrointestinal complaints (n/total)	Poor compliance (%)	Poor compliance (n/total)	Other (%)	Other (n/total)
Male donors								
40 mg iron	15.5	15/97	5.2	5/97	12.4	12/97	13.4	13/97
20 mg iron	18.8	18/96	6.3	6/96	16.7	16/96	3.1	3/96
0 mg iron (placebo)	20.8	20/96	6.3	6/96	21.9	21/96	11.5	11/96
Female donors								
40 mg iron	8.9	7/79	2.5	2/79	10.1	8/79	6.3	5/79
20 mg iron	20.3	16/79	6.3	5/79	11.4	9/79	6.3	5/79
0 mg iron (placebo)	24.1	19/79	3.8	3/79	10.1	8/79	11.4	9/79

TABLE 2. Serum ferritin concentration, number of donors with depleted iron stores (ferritin concentration <12 μg/L), and logarithm of the ratio of transferrin receptor to ferritin concentration (log[TfR/F]) for all donors with at least one follow-up visit

Visit number	Ferritin (μg/L) (mean ± SD)	Depleted iron stores (%)	Depleted iron stores (n/total)	log(TfR/F) (mean ± SD)
Male donors				
40 mg iron				
0	32.7 ± 27.5	26.3	20/76	1.54 ± 0.51
1	31.4 ± 18.8	16.2	12/74	1.47 ± 0.49
2	30.2 ± 20.8	17.9	10/56	1.50 ± 0.51
3	33.2 ± 26.7	13.0	7/54	1.52 ± 0.55
20 mg iron				
0	34.7 ± 36.3	25.3	20/79	1.48 ± 0.48
1	33.1 ± 33.3	21.8	17/78	1.46 ± 0.44
2	30.2 ± 32.7	25.4	15/59	1.47 ± 0.45
3	25.0 ± 19.8	24.5	13/53	1.52 ± 0.47
0 mg iron (placebo)				
0	35.1 ± 32.4	19.7	14/71	1.55 ± 0.50
1	27.5 ± 27.9	30.9	21/68	1.61 ± 0.45
2	24.9 ± 24.7	29.8	14/47	1.60 ± 0.52
3	21.4 ± 27.5	53.9	21/39	1.67 ± 0.53
Female donors				
40 mg iron				
0	19.3 ± 15.0	39.4	26/66	1.43 ± 0.65
1	28.5 ± 19.8	15.2	10/66	1.26 ± 0.49
2	31.4 ± 19.4	14.0	8/57	1.29 ± 0.54
20 mg iron				
0	20.0 ± 32.3	54.6	30/55	1.38 ± 0.46
1	23.3 ± 27.9	45.1	23/51	1.36 ± 0.42
2	23.5 ± 26.1	34.1	15/44	1.35 ± 0.49
0 mg iron (placebo)				
0	17.7 ± 15.0	48.9	23/47	1.39 ± 0.65
1	17.6 ± 14.5	44.2	19/43	1.40 ± 0.42
2	15.1 ± 12.3	48.7	19/39	1.55 ± 0.66

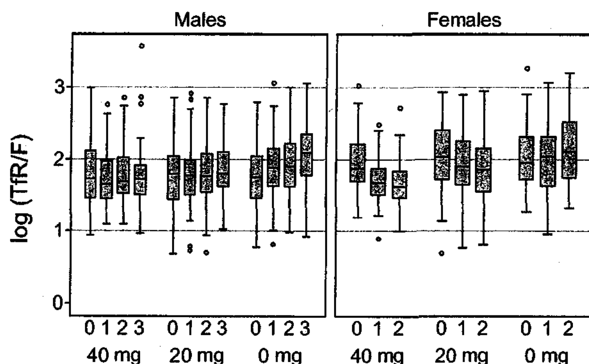

Fig. 3. Box-plots for the logarithm of the ratio of soluble transferrin receptor to ferritin concentration in male and female donors.

(Fig. 3), as was clearly demonstrated in the regression analysis (Table 3). The log(TfR/F) value increased by nearly 0.09 per donation in the placebo group, but changed only marginally in the two iron groups. Both iron groups differed significantly from the placebo group with respect to log(TfR/F).

Females

Of the 237 female volunteers (age range, 19-65 years) enrolled in the study, 96 (41%) dropped out, yielding a dropout rate of 28 percent in the 40 mg iron group, 44 percent in the 20 mg iron group, and 49 percent in the placebo group ($p = 0.015$; Fisher's exact test). A total of 69 (72%) of the female dropouts withdrew before their second visit (Table 1). The mean interval between visits was 88 days. Deferral from donation because of unacceptable dropout concentration values (<12.5 mg/dL) occurred in 13 of 546 visits (2.4%). This was the case more frequently in the placebo group than in the 20 mg and 40 mg iron groups (n = 10 vs. 2 vs. 1, $p = 0.001$; Fisher's exact test). Compliance was poor in roughly one-quarter of the female participants.

In the female placebo group, the mean concentration of serum ferritin decreased from 18 μg per L at baseline to 15 μg per L at the final visit, the number of females with depleted iron stores (ferritin <12 μg/L) remained constant (49%), and the mean soluble transferrin receptor concentration rose from 1.4 mg per L to 1.6 mg per L (Table 2, Fig. 4). In the female 20 mg iron group, serum ferritin increased from 20 μg per L to 24 μg per L, the number of individuals with depleted iron stores decreased from 55 percent to 34 percent, and the transferrin receptor concentration remained nearly constant (1.4 mg/L). In the female 40 mg iron group, ferritin concentration rose from 19 μg per L to 31 μg per L, transferrin receptor level fell slightly from 1.4 mg per L to 1.3 mg per L, and the number of individuals with iron depletion decreased from 39 percent to 14 percent.

The log(TfR/F) dropped in both iron groups, but rose continuously in the placebo group (Table 2, Fig. 3), as demonstrated by the regression analysis. The log(TfR/F) value increased by nearly 0.09 per donation in the placebo group (Table 3), but decreased by roughly 0.06 and 0.12, respectively, in the 20 mg and the 40 mg iron groups.

Side effects

Most donors (approx. 60%) did not report any side effects. There was no significant difference in the incidence of adverse effects between the three groups. In particular, the frequency of gastrointestinal complaints was low (11% in the 40 mg iron group, 13% in the 20 mg iron group, and 11% in the placebo group).

TABLE 3. Regression models for the change in log(TfR/F)

Predictor	Coefficient	95-percent confidence interval	p value
Male donors			
20 mg Fe^{2+}	−0.074	−0.121 to −0.028	0.002
40 mg Fe^{2+}	−0.118	−0.168 to −0.068	<0.001
Constant	0.091	0.058 to 0.123	<0.001
Female donors			
20 mg Fe^{2+}	−0.150	−0.238 to −0.061	0.001
40 mg Fe^{2+}	−0.209	−0.292 to −0.127	<0.001
Constant	0.086	0.018 to 0.153	0.012

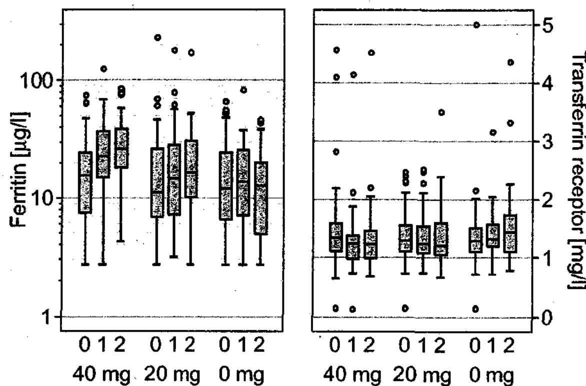

Fig. 4. Box-plot for the concentration of serum ferritin and soluble transferrin receptor in female donors.

DISCUSSION

Regular blood donation frequently leads to iron depletion, and it has been shown that iron supplementation can prevent this complication.[8,10,11] However, the exact dose needed to compensate for this type of iron loss remains unclear, and there is uncertainty as to whether iron supplementation is required in both male and female donors. Attempting to elucidate this complex issue more precisely, we monitored the logarithm of the TfR/F ratio as a measure of body storage iron in regular male and female whole-blood donors. The donors were randomly assigned to receive daily supplements containing selected vitamins plus 40 mg, 20 mg, or 0 mg of elemental iron. Dropout rates were marginally (male) or significantly (female) higher in the placebo group than in both iron groups. The reason for this finding is obscure.

Daily doses of 40 mg and 20 mg of elemental iron resulted in both a positive iron balance and an increase in storage iron in female donors and compensated for iron loss in males. This indicates that 20 mg of elemental iron per day is indeed sufficient to compensate for iron loss in both males and females. The differences in storage iron responses may be due to the shorter donation intervals in males (every 2 months) compared to females (every 3 months). It is likely that the ascorbic acid in the capsules may have increased the iron absorption by roughly 50 percent.[19] The question of whether the other vitamins may play any role in this context is speculative. The only reason for including these vitamins in the investigational products was our desire to improve the compliance rate.

In the present study, we monitored ferritin and soluble transferrin receptor levels as well as the logarithm of the TfR/F ratio. The latter variable, which was shown to have a highly linear correlation with body storage iron, is the most precise measure of body storage iron available.[14,15] Until now, body iron of blood donors was assessed mainly by measuring serum ferritin.[1,3,5-7] However, this variable is somewhat unspecific and may give false-high results in the presence of various underlying diseases.[2] In fact, if ferritin had been the only variable used for assessment of body storage iron, the effects of 20 mg elemental iron in males would have been underestimated in our study.

Interestingly, the number of side effects in the two groups treated with iron(II)-gluconate was only slightly higher than the number observed in the placebo group. In particular, the incidence of gastrointestinal side effects in the iron groups was very low (12%). Due to the slight risk of poisoning in children, iron capsules should be delivered in individual packages. Elemental iron preparations like carbonyl iron are preferred as an alternative by many experts due to the much higher lethal doses.[9,10,20,21] However, carbonyl iron is not available in the European countries. In comparison, bioavailabity of carbonyl iron is slightly lower than that of ferrous salts,[21] but side effects seem to be comparable: The incidence of gastrointestinal complaints for both preparations was reported much higher in two previous studies, probably due to the supplementation with higher doses of iron.[9,21] The utility of iron supplements for prevention of iron deficiency in menstruating female blood donors is currently being discussed.[20,22] However, others and we prefer a supplementation of iron for a short-term period after blood donation but not in general.

In conclusion, our results indicate that daily doses of 20 mg Fe^{2+} can adequately compensate for iron loss resulting from whole-blood donation in males who donate up to six times a year and in females who donate up to four times a year.

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2. 貧血と採血基準を考える
～血液学的立場から～

香川県赤十字血液センター
内田 立身

1．貧血の定義

貧血の定義について血液学の代表的な教科書をみると、①a reduction below normal in the concentration of hemoglobin or red blood cells in the blood[1] ②anemia is functionally best characterized by a hemoglobin concentration below normal[2] などの記載があり、健常人のヘモグロビンの下限値から判断するのが一般的である。米国人においては表1のような数字が用いられている[1][2][3][4]。この際、健常人として選ばれる対象のうち特に鉄欠乏状態の多い女性では血液学的に正常でない人が含まれ、下限域が低く算定される可能性があった。

表1　米国健常人のヘモグロビン(g/dL)下限値

	男性	女性	文献番号
WHO	13.0	12.0	3
Beutler E	14.0	12.3	1
Lee GR	13.2	11.6	2
NHANESⅢ	13.5	12.0	4

最近、Beutlerら[5]は米国人の貧血の定義としてNHANES-Ⅲ(The Third US National Health and Nutrition Examination Survey)[4]が行なったように、トランスフェリン飽和率16%以上、血清フェリチン10ng/mL以上の人を健常人として正常域の5%値未満を貧血としている(表2)。血液学的な貧血の定義として妥当な決め方である。

日本人の貧血の頻度について、私たちは「1981年～1991年」までの鉄欠乏の頻度を検索したことがあるが[6]、このデータをもとに鉄欠乏のない健常人を対象としてヘモグロビン値を求めたところ表3のとおりとなった。同じ方法で求められた斎藤ら[7]の成績とあわせると、鉄欠乏のない日本人のヘモグロビン下限値は男性12.8～13.2g/dL、女性11.8～12.1g/dLとなり、日本人成人の貧血の定義は男性13.0g/dL未満、女性12.0g/dL未満が妥当と考えられた。最近の日本人については鉄欠乏に関する正確なデータがなく、厚生労働省が行なっている「国民健康・栄養調査報告」などから鉄欠乏のない健常人のヘモグロビン値を求め、日本人の貧血の定義を定める必要がある。

表2　健常米国人のヘモグロビン(g/dL)下限値
(Beutler, 2006)

	男性(20～59歳)	女性(20～49歳)
白　人	13.7 (6,907人)	12.1 (2,966人)
アフリカ系	12.8 (434人)	11.1 (205人)

表3　鉄欠乏のない健常日本人のヘモグロビン値

	平均ヘモグロビン値	1標準偏差	5%正常分布値	文献
男性(284例)	14.8	1.0	12.8	6
女性(390例)	13.9	0.9	12.1	
男性(26例)	15.0	0.9	13.2	7
女性(134例)	13.4	0.8	11.8	

2．日本人の貧血の頻度

私たちは、1981～1991年にかけて3,015名の女性で貧血の調査を行なった。その成績は、健常者43.6%、貯蔵鉄欠乏33.4%、潜在性鉄欠乏8.4%、鉄欠乏性貧血8.5%、その他6.5%

表4　日本人の貧血の頻度(%)（平成16年度国民健康・栄養調査報告から）

年齢	男性			女性		
	平均Hb±SD	Fr<10(%)	Hb下限値	平均Hb±SD	Fr<10(%)	Hb下限値
20〜29	15.1±1.0	1.6	13.1	12.9±1.0	30.5	10.9
30〜39	15.1±0.8	1.2	13.5	12.7±1.2	36.5	10.3
40〜49	15.2±1.0	1.2	13.2	12.5±1.6	37.5	9.3
50〜59	14.9±1.2	1.8	12.5	13.2±1.1	10.0	11.0
60〜69	14.5±1.4	2.5	11.7	13.1±1.0	3.9	11.1
70≦	14.0±1.5	2.8	11.0	12.6±1.2	5.6	10.2
計	14.6±1.4	2.1	11.8	12.9±1.2	17.3	10.5

男性1,537名、女性2,634名の調査。

で40歳台前半では17.2%の鉄欠乏性貧血がみられた[6)]。

その後、日本人についての詳細なデータがなく、特に女性の鉄欠乏性貧血の頻度をみるには毎年厚生労働省が行なっている国民健康・栄養調査から類推するのがよいと思われる[8)]。表4はその成績である。高齢者を除くと男性の貧血は5.8%以下、鉄欠乏の頻度も2.5%以下であるが、女性は16.8%が貧血であり血清フェリチン低値(鉄欠乏)の頻度も高率であることから、ほとんどが鉄欠乏性貧血である。40歳台では25.0%に貧血があり同年代の半数（47.5%）が鉄欠乏状態にある。

また、香川県赤十字血液センターにおいて平成17年度に400mL献血を申し込んだ女性のうちヘモグロビン不足(Hb12.5g/dL未満)で献血ができなかった女性の比率[9)]を表5に示すが、30〜40歳台女性の約35%が献血できていない。また、日本赤十字社による全国的な調査によると[10)]、平成17年に比重不足で献血できなかった人は485,746人で、これは東京都で1年間に献血できた人の数407,235人をはるかに凌駕するほどである。

表5　ヘモグロビン不足で献血できない女性の割合
（平成17年：香川県赤十字血液センター）

年齢	Hb<12.5g/dL
16〜19	28.6%
20〜29	32.6%
30〜39	35.6%
40〜49	35.3%
50〜59	18.9%
60〜69	17.5%
全体平均	19.4%（申込者数 9,963人）

わが国の女性の貧血の頻度は欧米に比して高い。米国の国民健康・栄養調査報告によると、20〜40歳台の女性の鉄欠乏性貧血の頻度は5%、鉄欠乏状態は11%[11)]、米国24血液銀行における2003年度の女性ヘモグロビン不足(12.5g/dL未満)の割合は平均で6.6%(1.3〜13%)、Wisconsin州において17〜49歳では21〜23%である[12)]。わが国のこれに対応する成績は400ml献血ができなかった女性が該当し、16〜19歳で28.6%、20〜29歳で32.6%、30〜39歳で35.6%、40〜49歳で35.3%であり[13)]、どの調査をみても頻度は高いといわざるを得ない。

わが国で鉄欠乏の多い原因は鉄摂取量の不足にある。平成16年国民健康・栄養調査によると、男性の1日平均鉄摂取量は8.1mg、女性の1日平均は7.7mg（20〜39歳で6.9〜7.0mg)で必要量に比して少ない[8)]。日本人の必要鉄摂取量は男性10mg、月経のある女性12mgであるが、その差2mgは全血にして10〜12mLにしか相当せず、平均的月経量を30〜40mLとして外国並に15〜18mgは必要であろう。となるとわが国の月経のある女性は必要量の半分の鉄しか摂取していない。しかも鉄摂取量は過去の上記の調査によると年々減少してきている。

他方、米国における調査によると、白人男性で1日あたり17.2±0.3mg、女性で13.4±0.4mgで相当の開きがある[8)]。採血基準を考える際には、以上のようなわが国の事情を勘案して決める必要がある。

3．採血基準をどう決めるか

日本の現状を踏まえて、わが国の採血基準をどう決めたらよいかについて以下に私見をまじえて述べたい。

代表的な国の採血基準を表6に示す。このうちEU諸国とオーストラリアは男女差があるが、米国とわが国は男女差がない。わが国の採血基準は1986年に改定され、200mL献血と400mL献血に分け、比重法かヘモグロビン法で判定するようになっている。現在、貧血の定義はヘモグロビンで記載されており、わが国の医療機関のすべてがヘモグロビン法で貧血を診断しているので、ヘモグロビン法に統一することが望ましい。また献血も400mL献血が主流になりつつあるので諸外国に倣い200mL、400mLを一本化して表記するのがよいと考えられる。

表6　各国の採血基準（400mL相当）

	男性	女性
Council of EU	13.5	12.5
Australia	13.0	12.0
U.S.A	12.5	12.5
日本	12.5	12.5

1）　ヘモグロビンの正常範囲から決める

鉄欠乏のない健常者から正常分布域を定め、5％正常値を求めると男性13.0g/dL、女性12.0g/dLとなり、これ以上を採血基準とする方法はわかりやすく貧血の定義とも一致する。

2）　貧血状態にない人から採血する

赤血球は鉄欠乏の進展に伴い、小赤血球化、低色素性化する。図1、図2は男性および女性におけるヘモグロビンと赤血球恒数との関係で、MCV・MCHが低下するのは男性で12.5g/dL、女性で12.0～12.5g/dLである[14]。また、鉄欠乏性貧血82例の私達の検討から、ヘモグロビンの分布域の上限は13.0g/dLであることをみると、現行の米国やわが国の基準である12.5g/dLは矛盾しない数字となってくる。

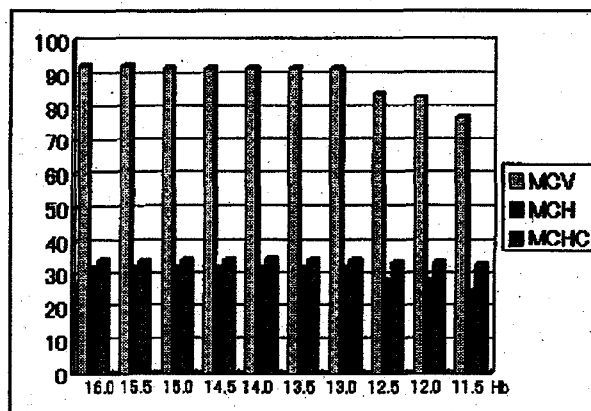

図1　赤血球恒数とヘモグロビン値の関係(男性)

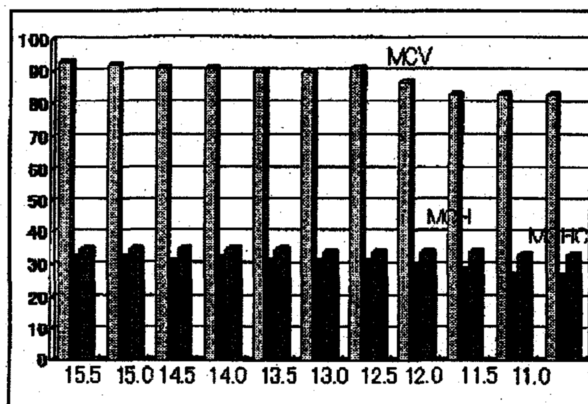

図2　赤血球恒数とヘモグロビン値の関係(女性)

3）　現在考えられる適切な採血基準は

上記を踏まえて採血基準について考察すると、わが国では鉄欠乏状態にある女性の頻度が高く、抜本的対策の見出せない現状では、貧血のない鉄欠乏からの採血をできるだけ避けるために女性の基準は12.0g/dLよりは12.5g/dLのほうが妥当と思われる。また、男性については貧血のない鉄欠乏はほとんどないが、12.5～13.0g/dLは貧血の人から採血することになり矛盾を生ずるので、13.0g/dLが妥当ではないかと思われる。

いずれにしても、採血基準の改定には正確なデータに基づく議論が必要である。それには、日本人の鉄欠乏性貧血、貧血のない鉄欠乏、鉄欠乏のない健常人の頻度（これは現行の国民健康・栄養調査の個々のデータから算出可能である）、献血申込者のヘモグロビン不足による男女別、年齢別不適格者の頻度などの解析によって決められるべきであろう。

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(17)

Mid-America Division
Badger-Hawkeye Region
Heart of America Region
Midwest Region
North Central Region

Dear Parent or Guardian,

Your 16-year-old has expressed an interest in donating blood at an upcoming American Red Cross blood drive. The states of Illinois, Iowa, Kansas, Nebraska, Minnesota, Missouri and Wisconsin allow 16-year-olds to donate blood with written parental/guardian consent. We are asking for your support by completing the attached consent form.

Please read the attached forms: "What You Must Know Before Giving Blood" and "What You Must Know About NAT – A New Blood Test." If you have any questions about the information contained in these documents, please call 1-800-448-3543 – M-F: 8 am - 9 pm, Sat: 9 am - 1 pm, Sun: 4 pm - 8 pm – and press Option 6 to speak to a Red Cross donor health consultant.

We support each student's willingness to give blood and ask that you offer your encouragement too. Much like voting and driving a car, the opportunity to donate blood and save a life has become a right of passage for thousands of high school students. Becoming a blood donor is a very personal decision, and we understand that parents and students may be somewhat apprehensive about taking this step. This is completely natural, so we want to provide you with some additional information about donating blood.

Blood donation is a safe procedure using single-use sterile needles and supplies. To ensure that your student has a positive experience, we recommend that they follow these guidelines:

- Get a good night's sleep before the blood drive.
- Eat well and drink plenty of fluids in the days leading up to the blood drive, especially the day of the drive.
- Drink at least 16 oz of caffeine free fluid (2 cups) 3-4 hours before the donation and after.
- Be honest and accurate about their weight (donors must weigh at least 110 lbs).

While the donation process is safe, reactions can occur. Most reactions are mild and can include fainting or small bruises. Our staff is fully trained to work with first-time and younger blood donors, and to respond to any reactions. We hope you will encourage your student to support our blood drive. Since one blood donation can be separated into three components, your student has the potential to save as many as three lives with a single donation.

Please note that the FDA requires that donors are asked specific questions about their health history. This information helps ensure the safety of the blood donor and the blood recipient. These questions are asked privately and are completely confidential.

You should be very proud of your son or daughter's decision to donate at the upcoming drive. *Please help support this act of generosity by completing the consent form prior to the drive.* If you are not currently a blood donor, please consider making an appointment for yourself. For more information call 1.800.GIVE.LIFE or visit our website at givebloodgivelife.org.

Sincerely,

David C. Mair MD

David C. Mair, M.D., Senior Medical Director

Form:
Informed Parental Consent for Persons Not of a Legal Majority

What this form is about

This form provides staff with a mechanism for documenting a parent or legal guardian's informed consent for someone not of legal majority to donate blood or blood components.

Who should use this form

This form applies to all staff who obtain informed special consent from donors or parent/legal guardian.

Instructions

- Ensure the region-identifying information is on the form.
- Instruct the parent/legal guardian to
  - Print the name of the son, daughter, or ward in the space provided.
  - Print his or her name.
  - Sign the consent form.
  - Date the consent form.
- Affix a Whole Blood Number/Donation Identification Number (WBN/DIN) to the form.

Revision History

Revision Number	Summary of Revisions
1.0	Initial version
1.1	Developed and released prior to revision history requirement
1.2	Revised instructions for completion of form Reformatted signature, date, and WBN lines

Informed Parental Consent for Persons Not of a Legal Majority

Information

This form must be completed by a parent or legal guardian for blood donations by any person who has not yet reached the age of legal majority as defined by the laws of the state in which the donor makes the blood donation.

Questions or concerns about the blood donation process should be directed to

Department: **Donor Health Consultants**

Phone Number: **(800) 448-3543 (Press Option 6)**

Hours of operation: **M-F: 8am-9pm, Sat: 9am-1pm, Sun 4-8pm**

Parental Consent

I have received and read a copy of "What You Must Know Before Giving Blood" describing the overall blood donation process.

I have received and read a copy of "What You Must Know About NAT- A New Blood Test" describing additional test procedures and any research-related attachments.

I understand that in the event it becomes necessary to notify my son, daughter, or ward of test results, the American Red Cross will send those results directly to my son, daughter, or ward.

I understand the information provided to me and have had an opportunity to ask questions about the information it contains. I hereby give permission for my son, daughter, or ward, to make a voluntary donation of blood to the American Red Cross during his or her legal minority.

A signed consent from the Parent/Guardian will be required for each donation until the donor reaches the age of majority.

Donor Name [son, daughter, or ward] (print) ______________________

Parent/Guardian Name (print) ______________________

Parent/Guardian *Signature* ______________________ **Date:** MM / DD / YY

WBN/DIN ➔

American Red Cross Blood Services
Washington, DC 20006

WHAT YOU MUST KNOW ABOUT NAT

Possible Use of Donor Information and Blood Samples in Medical Research

The American Red Cross Blood Services mission is to provide a safe and effective blood supply for patients who need blood transfusions. As part of this mission, the American Red Cross may conduct research. Some research is conducted with other institutions, such as academic centers and biomedical companies.

Some examples of the types of research are:

- Studies relating to testing, storing, collecting and processing blood to increase the safety of the blood supply.
- Studies of new test methods for infectious agents carried in the blood, like Nucleic Acid Testing (NAT).
- Studies of ways to recruit blood donors and to evaluate donor eligibility.

Participation does not require additional blood to be collected or additional time.

By signing your Blood Donation Record, you are giving consent to allow us to use a portion of your blood donation and donor information for research like that listed above. Donor information for research will not include anything that would identify you as the donor, such as your name or Social Security Number (SSN).

Confidentiality

American Red Cross policy requires protection of the confidentiality of your donor identifying information, results of tests on your blood samples and information collected at the time of donation. Strict procedures are observed at all blood collection facilities to maintain the confidentiality of donor information.

Your donor identifying information will not be released to other institutions for research purposes without your consent. Your age, gender, general geographic location, and test results may be used to evaluate important information about disease or donor recruitment, but this information is combined with information about other donors and not identified with you.

While study results may be published, donor names and other identifying information will not be revealed, except as required by law. Records are kept, as required by State and Federal Laws. The Food and Drug Administration (FDA) may need to review and copy donor records in order to verify study data. The FDA, however, is committed to protection of the confidentiality of donor identity.

Testing and Storage

Blood samples used by researchers are coded. This means that your donor identifying information, including name and SSN, is not used in connection with research. Coded samples can be linked to information about donors' identity only by authorized Red Cross personnel who are required to follow Red Cross procedures to maintain confidentiality.

Some of your sample or information may be saved for future research on viruses or other agents that may be carried in blood. Samples linked to your identifying information may be used, either

now or in the future, for infectious disease testing, as described in What You Must Know Before Giving Blood or in other information about a specific research study that is being conducted today. Your identified sample and information will not be used for genetic testing or for research unrelated to blood safety without your consent.

You will be notified in person, by phone, or by letter, about any test results that may impact your health. You will receive information about how these test results may affect your health and future eligibility as a blood donor.

Possible Participation in a Follow Up Study
If your test results are positive or unexpected, Red Cross staff may ask you to participate in a follow up study. Participation is voluntary and of no cost to you.

Benefits
By using new infectious disease tests like NAT, you may find out sooner if you are infected by one of the agents being tested. This may be important to your health.

Risks
There is a very low chance that your blood sample may give a false positive or true positive infectious disease result. If this happens, the blood that you donate will not be used for transfusion and there is the likelihood that you may not be able to donate again. If you are donating for a specific patient and have a positive test result, your blood donation will not be available for that patient. If you are donating blood for yourself and have a positive result, your blood donation may not be available to you.

Your Right Not To Participate
You may refuse to participate now or at any time during the donation process. If you decide that you do not want your donation or donor information to be used for possible research like that listed above, you will not be able to donate today. It is very important to include all donors in such research in order to provide a safe and effective blood supply.

If you decide not to participate at this time, your decision will not change your future relationship with the Red Cross.

If you begin donating and then decide that you do not want to participate, you must notify the blood collection staff before you leave the collection site. If you decide to withdraw in the future, contact the Scientific Support Office at (301) 212-2801. However, test information collected before your withdrawal may still be used or disclosed after your withdrawal.

Questions
If you have any questions about your donation, please feel free to ask the ARC staff member performing your confidential health history interview. If you have questions later, you can contact the Blood Center at 1-800-652-9742.

If you have scientific questions, you can call the Scientific Support Office at (301)212-2801. If you have any questions about your rights as a research participant, call the American Red Cross Institutional Review Board Administrator at (301)738-0630.

You have been given this information sheet to read and will be offered a copy to keep.

American Red Cross Blood Services
Washington, DC 20006

What You Must Know Before Giving Blood

Thank you for coming in today!

This information sheet explains how **YOU** can help us make the donation process safe for yourself and patients who might receive your blood. **PLEASE READ THIS INFORMATION BEFORE YOU DONATE!** You will be asked to sign a statement that says you understand and have read this information today. **If you have any questions now or anytime during the screening process, please ask blood center staff.**

Accuracy And Honesty Are Essential

Your **complete honesty** in answering all questions is very important for the safety of patients who receive your blood. We will ask you for identification each time you try to donate. Please register using the same identifying information each time you donate (name, date of birth, etc.). **All information you provide is confidential.** Although your interview will be private, it may require more than one American Red Cross employee to participate in or be present at your health history and blood donation.

What happens when you give blood

To determine if you are eligible to donate we will:

- ask questions about your health, travel, and medicines
- ask questions to see if you might be at risk for hepatitis, HIV, or AIDS
- take your blood pressure, temperature, and pulse, and
- take a small blood sample to make sure you are not anemic.

If you are able to donate we will:

- cleanse your arm with an antiseptic. **(If you are allergic to Iodine, please tell us!),** and
- use a new, sterile, disposable needle to collect your blood.

While you are donating: (the donation usually takes about 10 minutes)

- you may feel a brief "sting" from the needle at the beginning.

After donating we will give you

- a form with post-donation instructions, and
- a number to call if you have any problems or decide after you leave that your blood may not be safe to give to another person.

What to expect after donating

Although most people feel fine before and after donating blood, a small number of people may have a(n)

- lightheaded or dizzy feeling
- upset stomach
- black and blue mark, redness, or pain where the needle was, and
- very rarely, loss of consciousness, or nerve or artery damage.

We will give you a number to call to report any problems or concerns you may have following your donation.

Why we ask questions about sexual contact

Sexual contact may cause contagious diseases like HIV to get into the bloodstream and be spread through transfusions to someone else.

Definition of "sexual contact":

The words "have sexual contact with" and "sex" are used in some of the questions we will ask you, and apply to any of the following activities, whether or not a condom or other protection was used:

- vaginal sex (contact between penis and vagina)
- oral sex (mouth or tongue on someone's vagina, penis, or anus), and
- anal sex (contact between penis and anus).

Continued on back

American Red Cross Blood Services
Washington, DC 20006

What You Must Know Before Giving Blood, Continued

Persons who should not donate

You should not give blood if you

- had hepatitis on or after the age of 11
- had malaria in the past 3 years
- met any of the conditions listed in the CJD Information Sheet
- were held in a correctional facility (including jail, lock up, prison, or juvenile detention center) for more than 72 straight hours in the last 12 months.
- have had sexual contact in the past 12 months with anyone who is sick with hepatitis or AIDS
- had or were treated for syphilis or gonorrhea or tested positive for syphilis in the last 12 months
- were raped in the last 12 months
- **have AIDS or have ever had a positive HIV test**
  AIDS is caused by HIV. HIV is spread mainly through sexual contact with an infected person, or by sharing needles or syringes used for injecting drugs.
- **done something that puts you at risk for becoming infected with HIV**
  You are at risk for getting infected if you
  - have ever used needles to take drugs, steroids, or anything not prescribed by your doctor
  - are a male who has had sexual contact with another male, even once, since 1977
  - have ever taken money, drugs, or other payment for sex since 1977
  - have had sexual contact in the past 12 months with anyone described above
  - received clotting factor concentrates for a bleeding disorder such as hemophilia
  - were born in, or lived in, Cameroon, Central African Republic, Chad, Congo, Equatorial Guinea, Gabon, Niger, or Nigeria, since 1977.
  - since 1977, received a blood transfusion or medical treatment with a blood product in any of these countries, or
  - had sex with anyone who, since 1977, was born in or lived in any of these countries.
- have any of the following conditions that can be signs or symptoms of HIV/AIDS
  - unexplained weight loss (10 pounds or more in less than 2 months)
  - night sweats
  - blue or purple spots in your mouth or skin
  - white spots or unusual sores in your mouth
  - lumps in your neck, armpits, or groin, lasting longer than one month
  - diarrhea that won't go away
  - cough that won't go away and shortness of breath, or
  - fever higher than 100.5 F lasting more than 10 days.

Ineligible donors

We maintain a confidential list of people who may be at risk for spreading transfusion-transmitted diseases. By continuing this process, you consent to be entered in this confidential list of deferred donors if you are at risk for spreading such diseases. When required, we report donor information, including test results, to health departments, military medical commands, and regulatory agencies. Donation information may also be used confidentially for medical studies.

If you decide not to give blood

If you decide that you should not give blood, you may leave now.

Testing your blood

Your blood will be tested for hepatitis, HIV (the virus that causes AIDS), syphilis, and other factors. (There are unusual circumstances in which these tests cannot be performed.) You will be notified about test results that may disqualify you from donating blood in the future or that may show you are unhealthy. Your blood will not be used if it could make someone sick. (A sample of your blood or a portion of your donation might be used now or in the future for additional tests or other medical studies. Please tell us if you object.)

Though the tests we use are very good, they are not perfect. HIV antibodies may take weeks to develop after infection with the virus. If you were infected recently, you might have a negative test result, yet be able to infect someone. That is why you must not give blood if you are at risk of getting AIDS or other infectious diseases. **If you think you may be at risk for HIV/AIDS or want an HIV/AIDS test, please ask for information about other testing facilities. Please do not donate to get tested for HIV, hepatitis, or any other infections!**

Travel to or birth in other countries	Blood donor tests may not be available for some contagious diseases that are found only in certain countries. If you were born in, have lived in, or visited certain countries, you may not be eligible to donate.

Form:
CJD Information Sheet

What this form is about

This form explains Creutzfeldt-Jakob disease to the donor.

Who should use this form

This form applies to collections staff.

Revision History

Revision Number	Summary of Revisions
07/04	Developed and released prior to revision history requirement
05/08	• Removed watermark so sheet can be printed from eDOCs or eBinder • Revised American Red Cross Logo • Placed into System 3 Document template

CJD Information Sheet

Please <u>do not</u> donate if you—

- Since January 1, 1980 through December 31, 1996—
  - -Spent a <u>total time</u> that adds up to 3 months or more in any country(ies) in the United Kingdom (UK).
  - -The UK includes any of the countries listed in Table 1 below.
- Were a member of the U.S. military, a civilian military employee, or a dependent of a member of the U.S. military that spent a total time of 6 months on or associated with a military base in any of the following areas during the specified time frames—
  - -From 1980 through 1990 - Belgium, the Netherlands (Holland), or Germany
  - -From 1980 through 1996 - Spain, Portugal, Turkey, Italy, or Greece
- Since January 1, 1980 to present—
  - -Spent a <u>total time</u> that adds up to 5 years or more in Europe (includes time spent in the UK from
  - -1980 through 1996 and time associated with the military bases in Europe as outlined above).
  - -The European countries that are affected are listed below in **Table 1** and **Table 2**.
  - -Received a blood transfusion in any country(ies) listed in **Table 1** below.
  - -Received an injection of bovine (beef) insulin made in any of the countries listed below.
- Ever received—
  - -A dura mater (or brain covering) transplant during head or brain surgery.
  - -Human pituitary growth hormone (brain extract).
- Any blood relative has had Creutzfeldt-Jakob disease. A blood relative is your mother/father, grandparent, sibling, aunt/uncle, or children.
- Have been told that your family is at risk for Creutzfeldt-Jakob disease.

If any of these apply to you, your donation cannot be accepted. If you have any questions, please ask us. We sincerely appreciate your support.

Table 1

United Kingdom			
♦ Channel Islands	♦ Falkland Islands	♦ Isle of Man	♦ Scotland
♦ England	♦ Gibraltar	♦ Northern Ireland	♦ Wales

Table 2

Europe		
♦ Albania	♦ Hungary	♦ Poland
♦ Austria	♦ Ireland (Republic of)	♦ Portugal
♦ Belgium	♦ Italy	♦ Romania
♦ Bosnia/Herzegovina	♦ Kosovo (Federal Republic of Yugoslavia)	♦ Serbia (Federal Republic of Yugoslavia)
♦ Bulgaria		♦ Slovak Republic (Slovakia)
♦ Croatia	♦ Liechtenstein	♦ Slovenia
♦ Czech Republic	♦ Luxembourg	♦ Spain
♦ Denmark	♦ Macedonia	♦ Sweden
♦ Finland	♦ Montenegro (Federal Republic of Yugoslavia)	♦ Switzerland
♦ France		♦ Turkey
♦ Germany	♦ Netherlands (Holland)	♦ Yugoslavia (Federal Republic includes Kosovo, Montenegro, and Serbia)
♦ Greece	♦ Norway	

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American Red Cross Biomedical Services **Job Aid:** **Medication Deferral List**	Doc No 14.4.ja021	Version 1.1
	Approved by [signature] Quality Assurance	
	Approval date 05.04.06	

Please tell us if you are now taking or if you have EVER taken any of these medications:

- ☐ **Proscar® (finasteride) – usually given for prostate gland enlargement**
- ☐ **Avodart® (dutasteride) – usually given for prostate enlargement**
- ☐ **Propecia® (finasteride) – usually given for baldness**
- ☐ **Accutane®, Amnesteem®, Claravis®, or Sotret®, (isotretinoin) – usually given for severe acne**
- ☐ **Soriatane® (acitretin) – usually given for severe psoriasis**
- ☐ **Tegison® (etretinate) – usually given for severe psoriasis**
- ☐ **Growth Hormone from Human Pituitary Glands – used only until 1985, usually for children with delayed or impaired growth**
- ☐ **Insulin from Cows (Bovine, or Beef, Insulin) – used to treat diabetes**
- ☐ **Hepatitis B Immune Globulin – given following an exposure to hepatitis B**

  Note: This is different from the hepatitis B vaccine which is a series of 3 injections given over a 6 month period to prevent future infection from exposures to hepatitis B.
- ☐ **Unlicensed Vaccine – usually associated with a research protocol**

Please tell us if you are now taking or if you have taken any of these medications in the last 7 days:

- Clopidogrel
- Coumadin (warfarin)
- Heparin
- Plavix
- Ticlid
- Ticlopidine

IF YOU WOULD LIKE TO KNOW WHY THESE MEDICINES AFFECT YOU AS A BLOOD DONOR, PLEASE KEEP READING:

- If you have taken or are taking **Proscar, Avodart, Propecia, Accutane, Amnesteem, Claravis, Sotret, Soriatane, or Tegison**, these medications can cause birth defects. Your donated blood could contain high enough levels to damage the unborn baby if transfused to a pregnant woman. Once the medication has been cleared from your blood, you may donate again. Following the last dose, the deferral period is one month for **Proscar, Propecia, Accutane, Amnesteem, Claravis** or **Sotret**, six months for **Avodart** and three years for **Soriatane**. Tegison is a permanent deferral.
- **Growth hormone from human pituitary glands** was prescribed until 1985 for children with delayed or impaired growth. The hormone was obtained from human pituitary glands, which are found in the brain. Some people who took this hormone developed a rare nervous system condition called Creutzfeldt-Jakob Disease (CJD, for short). The deferral is permanent. CJD has not been associated with growth hormone preparations available since 1985.
- CJD has been reported in extremely rare cases in Australian women who took **gonadotropin from human pituitary glands** for treatment for infertility. Gonadotropin from human pituitary glands was manufactured and distributed outside the United States and was never marketed in the United States to treat infertility. Human chorionic gonadotropin which is used for fertility treatments in the United States is not derived from human pituitary glands and is not a cause for deferral.
- **Insulin from cows (bovine, or beef, insulin)** is an injected material used to treat diabetes. If this insulin was imported into the US from countries in which "Mad Cow Disease" has been found, it could contain material from infected cattle. There is concern that "Mad Cow Disease" is transmitted by transfusion. The deferral is indefinite.
- **Hepatitis B Immune Globulin (HBIG)** is an injected material used to prevent infection following an exposure to hepatitis B. HBIG does not prevent hepatitis B infection in every case, therefore persons who have received HBIG must wait 12 months to donate blood to be sure they were not infected since hepatitis B can be transmitted through transfusion to a patient.
- **Unlicensed Vaccine** is usually associated with a research protocol and the effect on blood transmission is unknown. The deferral is for one year.
- If you have taken **Clopidogrel, Plavix Ticlid, or Ticlopidine in the last 7 days**, these medications affect the portion of your blood called platelets. If you are donating platelets, your donated blood could contain high enough levels of the medications that it could affect the quality of the platelets that you give. Once the medication has been cleared from your blood, you may donate platelets again. Following the last dose, the deferral period is 7 days.
- If you have taken **Coumadin (Warfarin) or Heparin in the last 7 days**, these medications can affect the blood's ability to clot, which might cause excessive bruising or bleeding when you donate. Therefore, we ask that you be off of these drugs for 7 days prior to giving blood. Following the last dose, the deferral period is 7 days.

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American Red Cross Biomedical Services Washington, DC 20006	**Controlled Documents Approval**	

SECTION 1: Document Package Information

Transmittal Sheet Title: Revised health History Tables and Related Douments Number: 2522
Version: 1.0

Document Title: Time Period: May 2006

List Documents Here:	14.4.ja041, v-1.1	14.4.tbl010, v-1.4
14.3.019, v-1.3	14.4.ja049, v-1.2	14.4.tbl011, v-1.2
14.3.070, v-1.2	14.4.tbl001, v-1.3	14.4.tbl012, v-1.2
14.3.092, v-1.2	14.4.tbl002, v-1.3	14.4.tbl016, v-1.3
14.3.094, v-1.2	14.4.tbl003, v-1.4	14.4.tbl021, v-1.2
14.4.ja021, v-1.1	14.4.tbl004, v-1.4	14.4.tbl023, v-1.3
14.4.ja028, v-1.2	14.4.tbl005, v-1.2	14.4.tbl024, v-1.3
14.4.ja029, v-1.4	14.4.tbl006, v-1.3	14.4.tbl025, v-1.2
14.4.ja031, v-1.3	14.4.tbl008, v-1.2	14.4.tbl026, v-1.2
14.4.ja032, v-1.1	14.4.tbl009, v-1.3	
List Documents Here (continued):		
14.4.tbl027, v-1.2	14.4.tbl039, v-1.3	
14.4.tbl028, v-1.2	14.4.tbl044, v-1.3	
14.4.tbl029, v-1.4	14.4.tbl045, v-13.	
14.4.tbl030, v-1.2	14.4.tbl046, v-1.3	
14.4.tbl031, v-1.3	14.4.tbl047, v-1.3	
14.4.tbl033, v-1.2	14.4.tbl048, v-1.3	
14.4.tbl034, v-1.4	14.4.tbl201, v-1.1	
14.4.tbl035, v-1.2	14.4.tbl202, v-1.1	
14.4.tbl036, v-1.2	14.4.tbl208, v-1.1	

SECTION 2: Approvals

Your approval signifies that you have reviewed the documents according to the requirements for your functional area.

Signatory Name	Role		Signature	Date
Please print or type name here	*Check role*			
Pat Demaris	Process Owner	☒		
	CEO/ Division VP	☐	Pat Demaris	05/05/06
	None	☐		
Anne Eder	Medical Office	☒	Anne Eder	05/05/06
	None	☐		
	Executive QA	☒		
	System QA	☐		
	BIT-QRM	☐		
	Testing Support QA	☐		
	Facility Quality Director	☐	[illegible signature]	05.05.06

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(18)

平成１５年度　厚生労働科学研究費補助金（医薬品等医療技術リスク評価研究事業）
分担研究報告書

4．採血により献血者に起こる副作用・合併症の解析

―平成14年の全国データから―

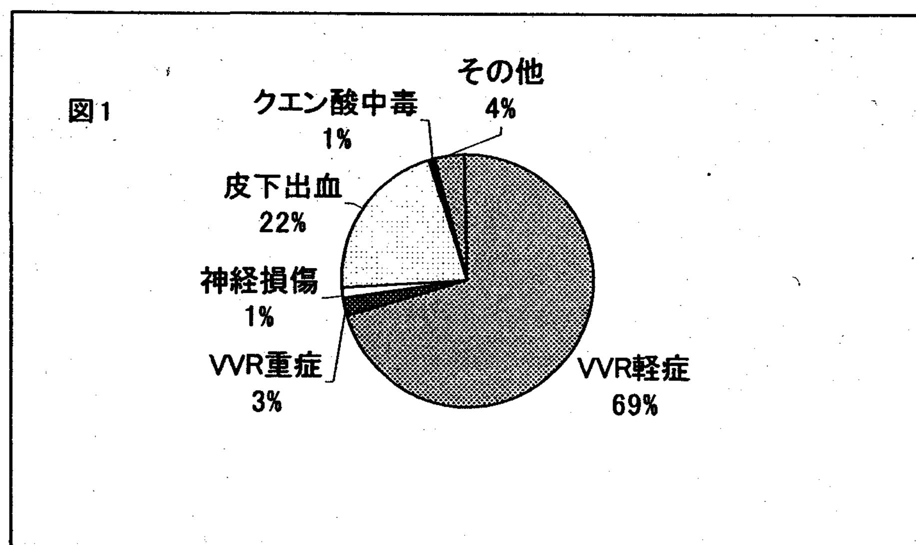
分担研究者
佐竹　正博　（東京都赤十字血液センター）
中村　榮一　（東京都赤十字血液センター）

日本赤十字社では、献血時の採血によって献血者に起こる副作用や合併症のデータを集積しているが、ここでは全国の血液センターから集められた平成14年のデータをもとに解析を試みた。

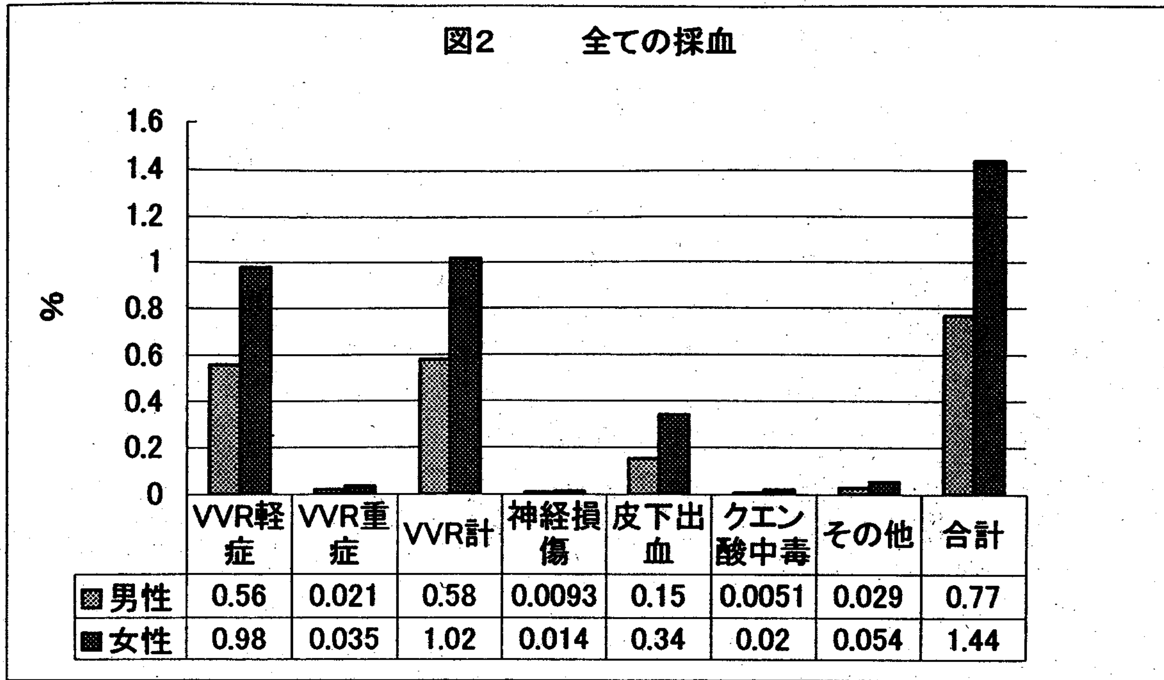
まず、すべての採血種における全献血者の副作用の頻度を表に示した。

	VVR 軽症	VVR 重症	神経損傷	皮下出血	クエン酸中毒	その他	合計
%	0.73	0.026	0.011	0.23	0.011	0.039	1.04

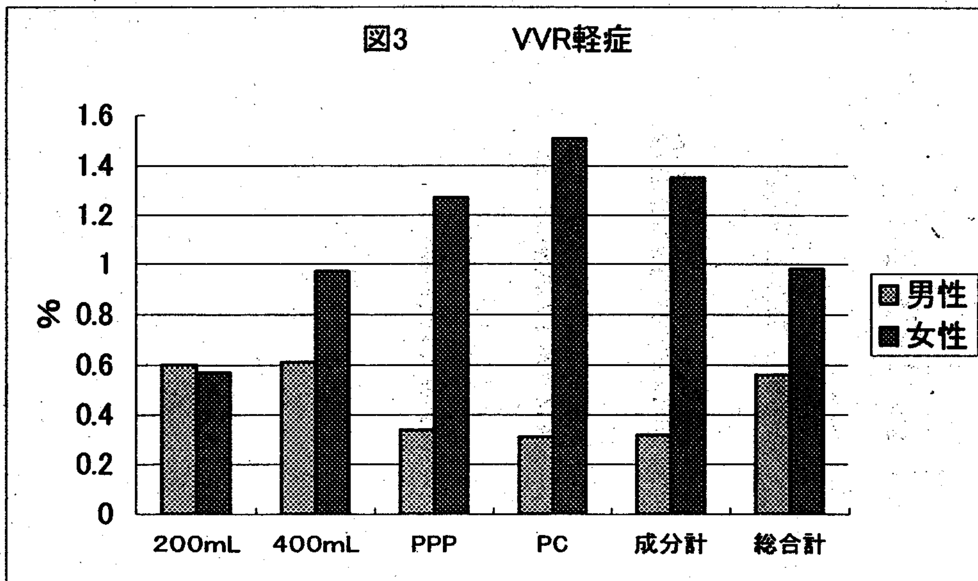
全献血者の約1%に何らかの副作用が起こっており、その73%はVVR（vasovagal reaction、血管迷走神経反応）である。献血者に長期にわたる愁訴・運動障害などを起こす可能性のある神経損傷が1万人に1.1人の確率で起こることは重大である。副作用の割合を示したのが図1である。VVRに次いで、皮下出血が22%を占めている。



これを男女別にみたのが次の図2である。



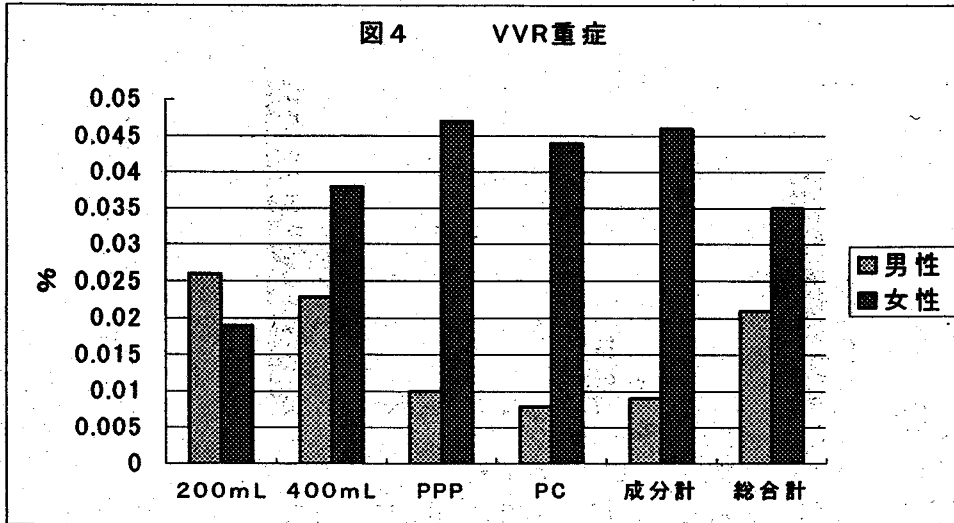
男女別でとくにパターンの大きな変化はないが、すべての副作用において女性のほうがその頻度が高い。しかしながら、これを採血種別にみていくと男女間でかなり大きな差があることがわかる。図 3 は比較的軽症の VVR の発生頻度を採血種別にみたものである。



200mL 採血では男女ほぼ同じ頻度で VVR が起こっているが、400mL になると女性のほうが有意に多くなる。これは、女性のほうが一般に循環血液量が少なく、血管内の volume loss による症状が現れやすく、それが VVR に加算されて頻度が高くなったものと思われる。PC や PPP の成分採血になると、男性ではむしろ VVR が少なくなっているのに対し、女性ではさらに頻度が高くなっている。女性で多くなるのは、前述のように血漿採取量の増加の影響が出ているものと思われるが、男性でかえって少なくなる理由は不明である。男性の場合、血漿採取量が循環血液量に影響を及ぼさない範囲では、専用椅子に 1 時間近くゆっくり座って採血を受ける成分採血の方が心理

的に余裕があり、VVR が起こりにくいこともあるのではないかと想像される。

重症の VVR では図 4 にように 200mL 採血ではむしろ男性の方が多い。成分採血では女性は男性の 5 倍ほど重大



な転帰をとりやすい。男女とも 200mL 採血では循環血液量に影響が出ることはほとんど考えられないので、この採血において男女の VVR の頻度がほぼ同じであることは、純粋に神経学的な機序のみで起こる VVR の頻度に性差はあまりないことを示すものといえる。図 5 は軽症と重症を合わせた全 VVR の頻度である。

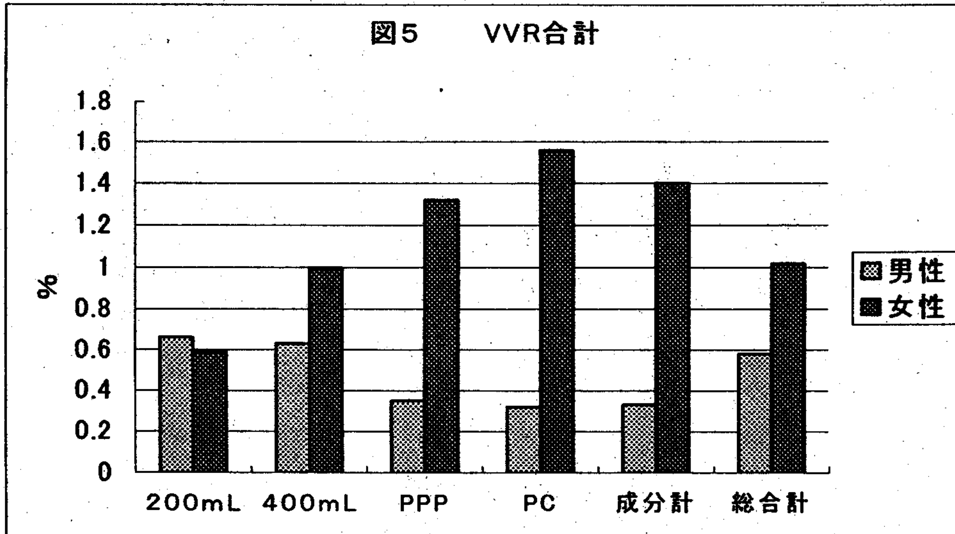


図6は神経損傷の頻度である。ここでは予想されるように採血種別による頻度の差はほとんどない。これはいっぽうでこのデータ収集が大きな片寄りのないものである事を示すものと思う。女性のほうがどの採血種別でも男性より頻度が高い。女性はより痛みに敏感であることが影響していると思われる。これは RSD(reflex sympathetic dystrophy)などが女性に多いといわれる事などからも推察される。

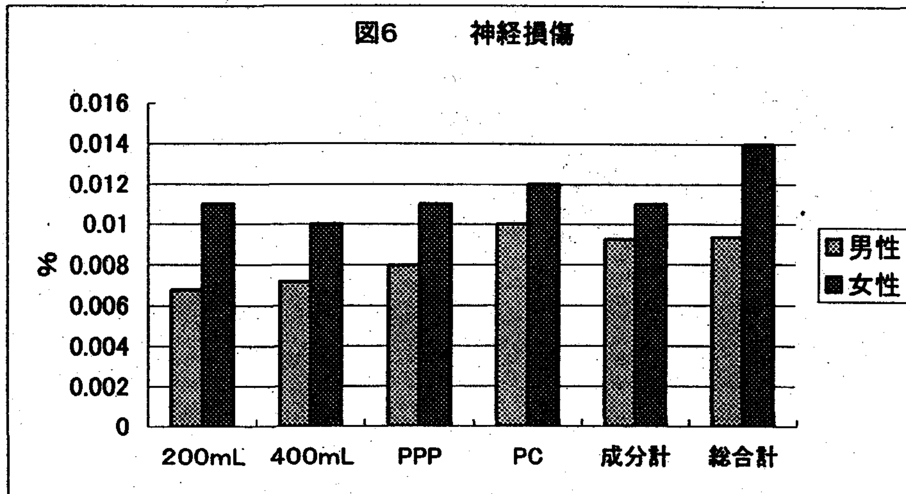


図7は皮下出血の頻度である。特徴的なのは、200mL、400mL 採血ではどちらも同程度に頻度が低いのに対し、成分採血では約6倍ぐらい高いことである。これは、穿刺針が長時間静脈内に留置されている間に血管壁を傷つける可能性が高いためであると考えられるが、さらに、長時間異物が挿入されていることにより、創傷の治癒機転が少なからず阻害される事もあるのではないかと考えられる。どの採血種でも女性は男性のちょうど2倍の報告がある。女性の方が美容上より気にしやすいこともあるだろうが、破綻血管からの止血について女性が本質的に弱点を持っている可能性はないだろうか。

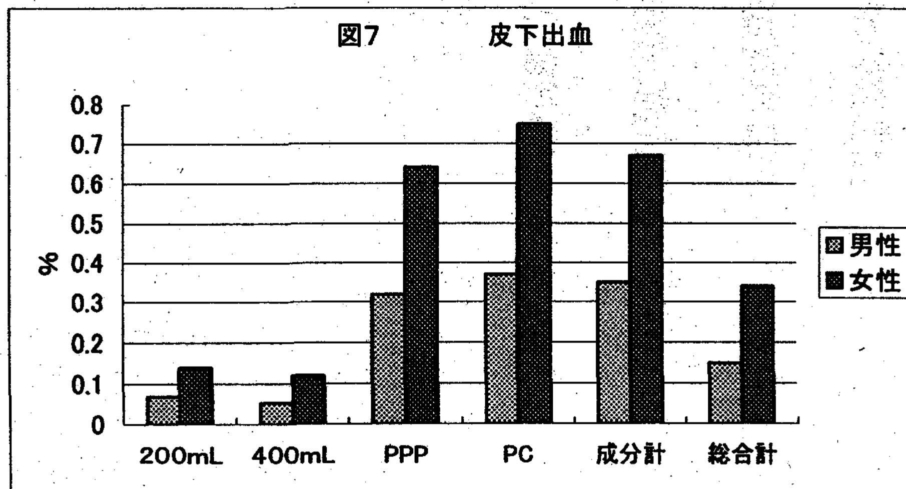


図8はクエン酸中毒の頻度で、母集団は成分採血者のみとした。血漿採血（PPP）よりも血小板採血（PC）の方が遥かにクエン酸中毒を起こしやすい。これは採取血小板の凝集を防ぐためにPC採取の場合はACD輸注比を高く設定するためと、PC 採取の方が時間が長くかかるためと思われる。また、女性の方が圧倒的に頻度が高いのは、体格が小さいために循環血液量が少なく、クエン酸の血中濃度が高くなりやすいためと思われる。

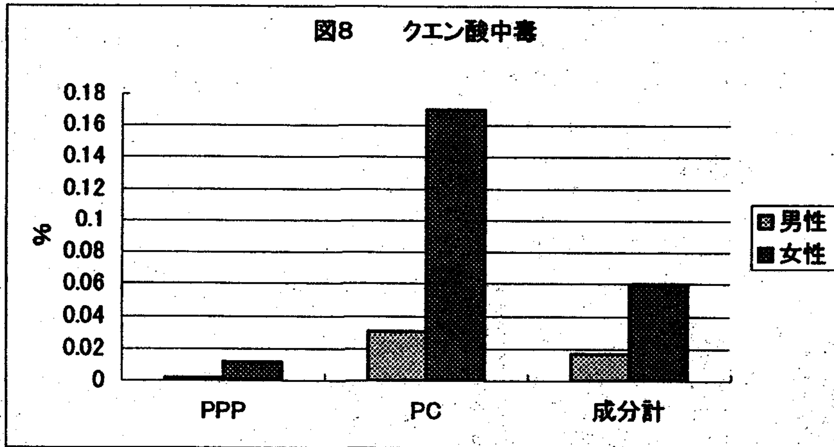


図9はその他の副作用である。

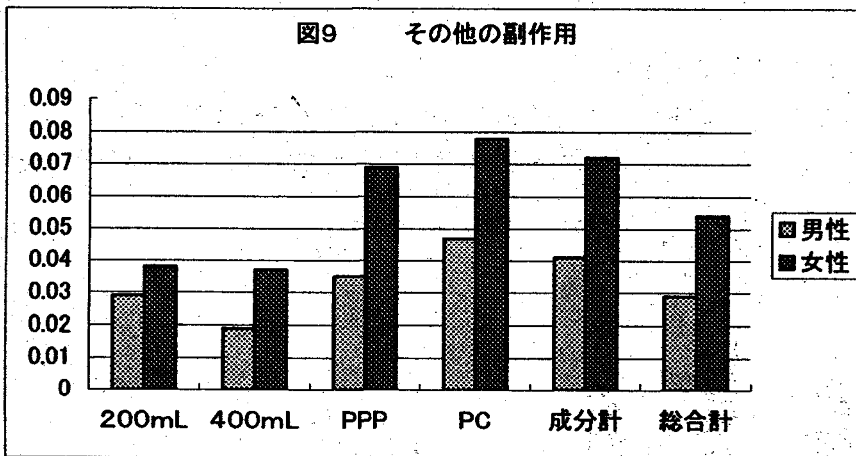
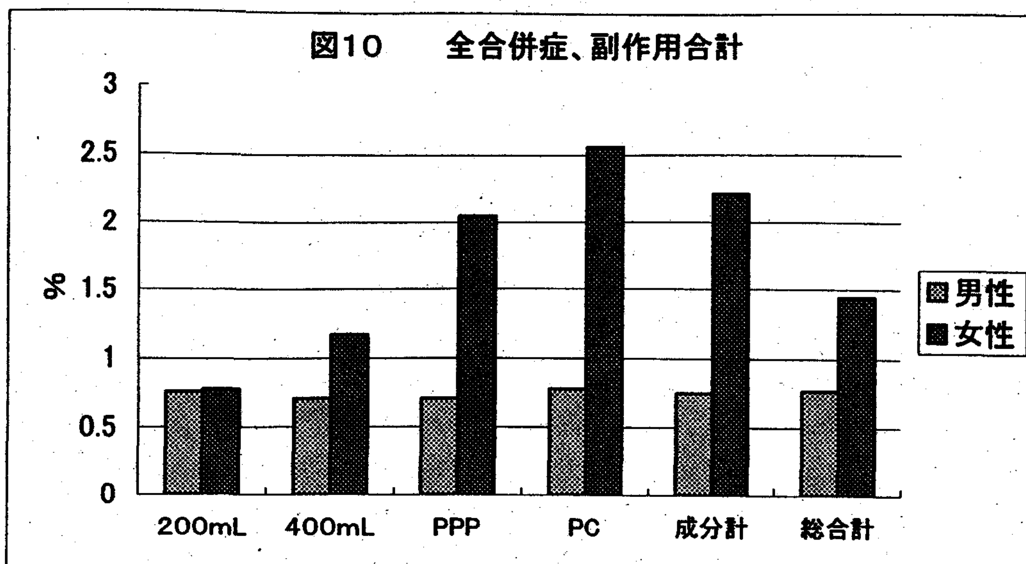
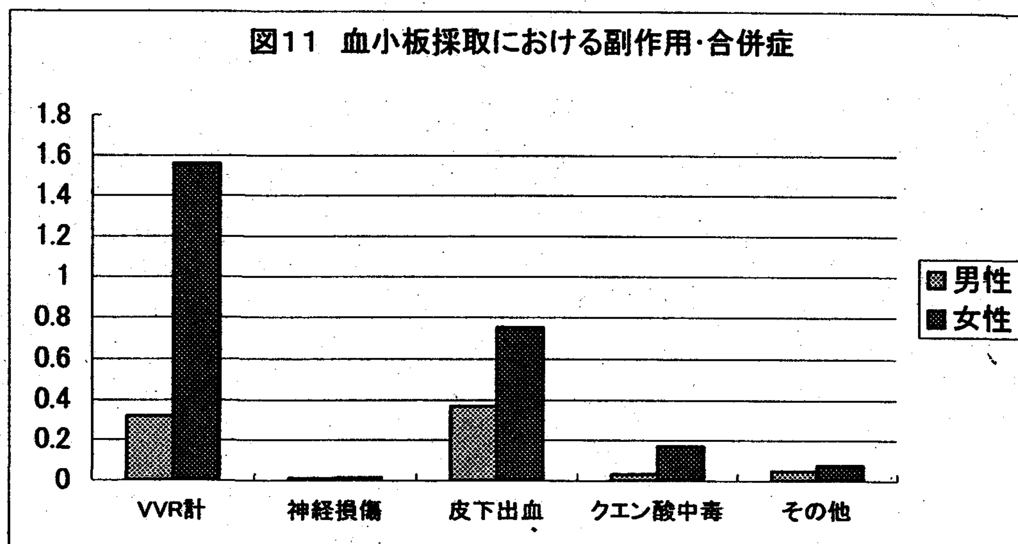


図10は、すべての採血副作用・合併症の合計の頻度を採血種別、男女別に合計したものである。おもしろいことに、男性ではすべての採血種でほぼ同じ合併症頻度を示す。これに対し女性では、200mL、400mL、PPP、PC の順に直線的に頻度が高まっていく。これに最も寄与しているのが VVR で、以下皮下出血、クエン酸中毒と続く。女性の PC 採血者において 2.5%もの献血者に副作用が出ている事実は注目されなければならない。血小板採取で起こる副作用をまとめると図11のようになり、女性においては VVR、次に皮下出血の順となる。成分採血後の止血法については改善の余地がある。





まとめ

全献血者の約1%に何らかの副作用･合併症が起こる。その73%はVVRであり、皮下出血が22%である。女性は男性の1.87倍合併症が起こりやすい。採血種別では、PC採血において最も頻度が高く、PPP、400mLと続く。これは女性にのみ認められる現象で、男性ではどの採血種別でも同じ頻度である。女性でこの頻度を高くしているのがVVR、次いで皮下出血である。

男性において、採血の環境･状況が異なるどの採血種でも頻度が同じであり、また200mL採血では男女の差はまったくないことは、この頻度が日本で不可避的に起こる採血合併症の頻度ではないかということを示唆する。いっぽう、女性での頻度の増加分は採血状況の何らかの改善によって防ぎうるものではないかということも示唆す

る。最も問題となるのはおそらく循環血液量に対する採血量の過重な負担であろう。現行の採血量・採漿量は、献血を継続していっても貧血に陥らない量、また急速脱血しても循環動態に影響を与えない量（循環血液量の12～13%）を基準に決められている。後者のよりどころとなるのは、健常者が安定した状態にあって脱血した場合のデータであると思われる。生理学的研究においてはこれは間違いのないデータであろうが、献血の場合は、問診において全身状態に問題のある献血者をお断りしているとはいえ、脱水や睡眠不足などあらゆる全身状態の献血者が採血を受け得る状況にある。このような献血者群から400mL以上採血した場合は、失神などの副作用は容易に起こるであろうと思われる。PC、PPP、400mL採血でのVVR増加分がこのような献血者群でのVVRの増加によるものかどうかについてはデータはないが、その可能性は十分にあると思われる。

十分に検討された現行の基準で採血を行っても１%もの献血者にVVRなどが起こっている。日赤の血液センターでは、これらの副作用を少しでも少なくするために、採血前後の水分補給、採血後の十分な休息、退出後の過ごし方での注意点の周知などに努めている。そして今回まとめられたデータをもとに、さらにどのような対策が適切であるかを現在検討中である。将来、献血時の採血量を増やす場合には、性差、体重、循環血液量、採血種別について十分に検討する必要がある。とくに女性での採血量については慎重に検討しなければならない。女性でのPC、PPP、400mL以上の採血では何らかの新たな基準が必要であろう。問診でのドナー選択と献血前後のドナーの処置法も再検討しなければならない。1年間に600万人の献血者から採血している状況から得られたデータは、小数の実験・麻酔例からのデータより重いものがあるのではないだろうか。

19

[原著]

血管迷走神経反応の予防の試み ―ハイリスクドナーに休憩と水分摂取を勧めるパンフレットを渡したことの効果

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Trial prevention of vasovagal reaction —The effect of handing pamphlets to high risk donors instructing them to take rest and drink water

Saitama Red Cross Blood Center
Yukiko Kaga, Taeko Nukita, Machiko Arakawa,
Toshiaki Shibasaki, Kenichi Yamazaki and Hideaki Mizoguchi

抄 録

血管迷走神経反応(VVR)は献血者の副作用として一番多く，献血者の約1%に起こる。VVRを起こしやすい献血者のグループ(ハイリスクグループ)があることが知られている。

我々はVVRの頻度を減らす目的で，ハイリスクグループのうち①全血献血の初回の若年(10歳代と20歳代)の男女，②成分献血の中高年(50歳代と60歳代)の女性に対し，①休憩を30分以上取ること，②水分摂取をすることを勧めるパンフレットを手渡した。

その結果，パンフレットを渡すようになった2004年度と2005年度ではそれ以前の2002年度と2003年度に比し月ごとのVVRの頻度は低下した。2003年度と2004年度を比較すると軽症のVVRは男女とも低下した。重症のVVRは男性では低下しないが，女性では全体でも有意に低下し，血漿献血と400mL献血で有意に低下した。この方策は，VVRの減少に有効な方法と考えるが，若年男性の重症に対しては他の方策を考える必要がある。

Abstract

Among adverse events related to blood donation, vasovagal reaction (VVR) occurs most frequently and its incidence comprises around 1% of donors. It is well known that there are high risk populations who are susceptible to VVR.

In order to decrease the incidence of VVR, we prepared pamphlets that instruct donors to take rest for at least 30 minutes and to drink water after blood donation, and handed these pamphlets to 2 high risk group donors: first-time

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young whole blood donors and middle aged apheresis female donors. As a result, the incidence of VVR decreased after handing the pamphlets to high risk donors. Comparing the incidence of VVR before and after handing the pamphlets to donors, mild VVR decreased in both male and female donors. As far as the incidence of severe VVR is concerned, the incidence of VVR among male donors did not change, though the incidence of VVR among female 400mL whole blood donors and plasma apheresis donors decreased significantly. The pamphlets that we prepared effectively decreased the incidence of VVR but we must consider other methods of decreasing the incidence of severe VVR among young male donors.

Key words: blood donation, vasovagal reaction, rest, water intake

はじめに

献血後の副作用は献血者の約1%に起こることが知られている[1]。その主なものは血管迷走神経反応(vasovagal reaction, VVR)，神経損傷と皮下出血である。VVRは全副作用のうち約75%を占める。VVRは転倒の原因となり，重篤な副作用に繋がる可能性がある。VVRによる転倒は全国で，年間100～150人の献血者に起こり，大きな問題と考える[2)~4)]。転倒事故を少なくするためにはVVRの発生率を下げる努力と転倒の直接的な予防策を立てる必要がある。

全血献血でVVRを起こしやすい人々は，①初回，②低体重，③若年，④白人，⑤若年初回の献血者では女性と報告されている[5)~7)]。一方，成分献血では①循環血液量の少ない人，②中高年の女性，③サイクル数の多い人等が挙げられる[8)]。埼玉県の予備的な調査でも同様の傾向がみられ，中高年の女性の成分献血ではVVRが1時間以上持続する例が多い。

今回，VVRの発生率を低下させる目的で，VVRのリスクの高い献血者に対し，図1に示すような献血後に①30分以上の休憩することと，②水分摂取を勧めるパンフレットを渡し，そのVVR発生に対する効果を検討した。また同時に口答でもその内容を献血者に話すようにした。

方法と対象

対象とした献血者は2004年5月から2005年4月までの1年間に埼玉血液センターに来訪した献血者243,182人(男性149,271人，女性93,911人，全血献血159,186人，成分献血83,996人)であった(表

看護師からのお願い

- 採血終了後、少なくとも30分休息してください。
- 水分を補給してください。
- 内出血の予防のため、15分間は止血バンドをしてください。
- 針痕をもんだり、こすったりしないでください。

図1　VVRのハイリスクの献血者に渡すパンフレット

看護師からのお願い

- 採血終了後、少なくとも15分休息してください。
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- 針痕をもんだり、こすったりしないでください。

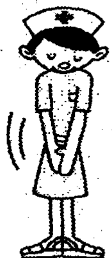

図2　VVRのローリスクの献血者に渡すパンフレット

1）。それらの献血者のうち，VVRのリスクが高いとされる初回の若年（10歳代と20歳代）の男女で全血献血をした人と再来の中高年（50歳代と60歳代）の女性で成分献血をした人に2004年5月から図1に示すようなパンフレットを渡した。その内容は献血後に①少なくとも30分以上は休憩することと，②水分摂取をすることを勧める内容である。それ以外の献血者に対しては図2に示すようなパンフレットを渡した。その内容の主なものは①少なくとも15分以上休憩すること，②水分摂取を勧める内容である。

パンフレットを渡し始めたのが，2004年5月であるので，年度の区切りを5月から次年度の4月までとした。つまり，2004年5月から2005年4月を2004年度とし，その月ごとのVVRの発生頻度とそれ以前の2002年度および2003年度の月ごとのVVRの発生頻度と比較した。2005年度の月ごとのVVR発生頻度も調べ比較した。

さらに，2003年度と2004年度のVVRの発生頻度についてその効果を男女別，献血の種類別，VVRの重症度別に比較検討した。

なお，比較の対照とした2003年度の献血者は総献血者数246,056人（男性149,898人，女性96,158人，全血献血161,757人，成分献血84,299人）であった（表2）。

VVRの重症と軽症の分類は表3に示すように，日本赤十字社標準作業手順書に準拠した[9]。つまり，軽症では気分不良，顔面蒼白，あくび，冷汗，悪心，嘔吐，5秒以内の意識喪失であり，重症になると，これらの症状に加え，5秒以上の意識喪失，けいれん，尿失禁，脱糞などが起こる。身体所見としては血圧の低下と徐脈，呼吸数の低下などがみられ，この重症例の一部に転倒例が含まれる。

結　果

図1あるいは図2のパンフレットを渡すようになった2004年度（パンフレットを渡すようになった2004年5月から2005年4月までとする）の各月のVVRの頻度は2002年度あるいは2003年度の各月のVVRの頻度に比し低い値を示した（図3）。つまり，2002年度と2003年度の各月のVVRの発生頻度はほとんどの月で1%を超えていたが，パンフ

表1　埼玉県赤十字血液センターにおける2004年度の献血者数

	全血献血		成分献血		総計
	200mL	400mL	PC＋PPP	PPP	
男性	13,595	85,369	21,009	29,298	149,271
小計	98,964		50,307		
女性	36,910	23,312	8,243	25,446	93,911
小計	60,222		33,689		
総計	159,186		83,996		243,182

200mL：200mLの全血献血
400mL：400mLの全血献血
PC＋PPP：血小板献血，PPP：血漿献血

表2　埼玉県赤十字血液センターにおける2003年度の献血者数

	全血献血		成分献血		総計
	200mL	400mL	PC＋PPP	PPP	
男性	14,328	85,420	19,676	30,474	149,898
小計	99,748		50,150		
女性	37,138	24,871	7,946	26,203	96,158
小計	62,009		34,149		
総計	161,757		84,299		246,056

200mL：200mLの全血献血
400mL：400mLの全血献血
PC＋PPP：血小板献血，PPP：血漿献血

表3　VVRの重症度分類[9]

分類	症　状	血圧（max，mmHg）採血前→測定最低値	脈拍数（/分）採血前→測定最低値	呼吸数（/分）
軽症	気分不良，顔面蒼白，あくび，冷汗，悪心，嘔吐，意識消失（5秒以内），四肢皮膚の冷汗	120以上→80以上 119以下→70以上	60以上→40以上 59以下→30以上	10以上
重症	軽度の症状に加え，意識喪失（5秒以上），けいれん，尿失禁，脱糞	120以上→79以下 119以下→69以下	60以上→39以下 59以下→29以下	9以下

レットを渡すようになった2004年度の各月のVVRの発生頻度は1%未満となり，同様のVVRの低下傾向は2005年度でも持続していた。

男性の軽症のVVRの頻度は2004年度の発生率の方が2003年度の発生率に比し，全体で有意に低下した（図4）。軽症が大部分を占めるので，献血者全体でも有意に低下した。まずその献血の種類による違いをみると，血漿献血，血小板献血，400mLの全血献血，200mL全血献血のいずれでも有意に低下した（図4）。

女性の軽症のVVRの頻度は，2004年度の発生率は2003年度の発生率に比し，全体で有意に低下した（図5）。また，その献血の種類による違いをみると，血漿献血，400mL献血，200mL献血で有意に頻度が低下した（図5）。しかし，血小板献血では有意の頻度の低下は認められなかった。

男性の重症例で調べると，その頻度は2003年度も2004年度も0.03%と軽症例がそれぞれ0.7%と0.5%であるのに比べて，約1/10と少なかった。2004年度のVVRの発生率は2003年度の発生率と有意の差を認めなかった（図6）。また，いずれの献血種別でも差を認めなかった。とくに，200mL

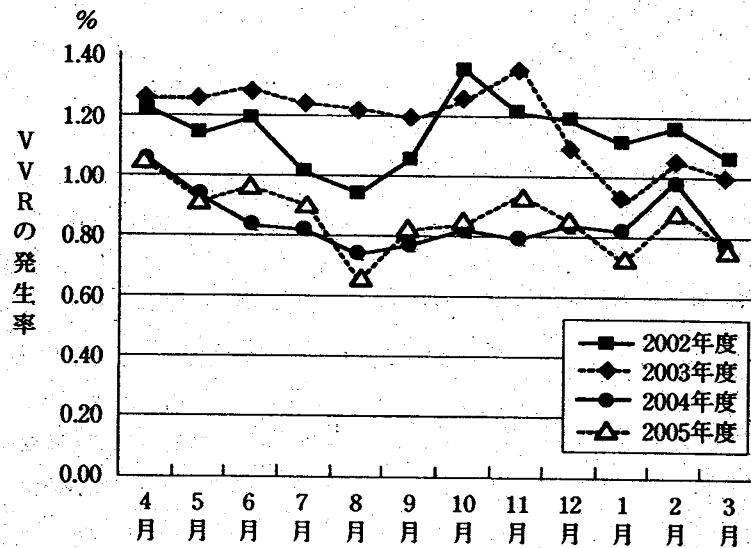

図3　VVRの発生率―2002年度，2003年度，2004年度，2005年度の月ごとのVVR発生率

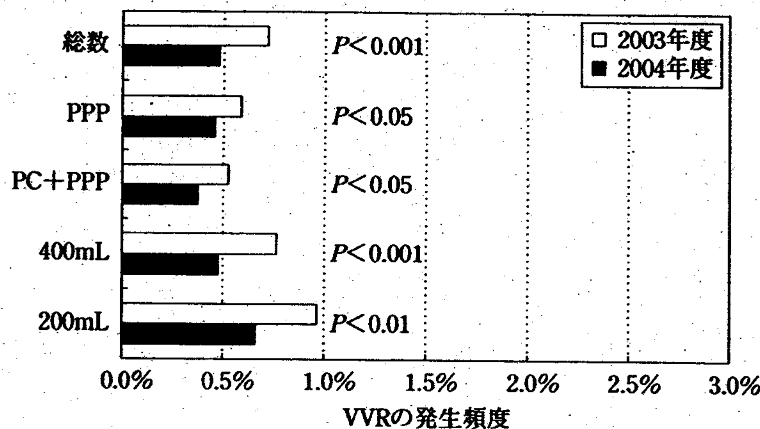

図4　埼玉赤十字血液センターにおける男性の軽症VVRの発生頻度の年度別の比較
PPP：血漿献血，PC＋PPP：血小板献血　400mL：400mLの全血献血　200mL：200mLの全血献血

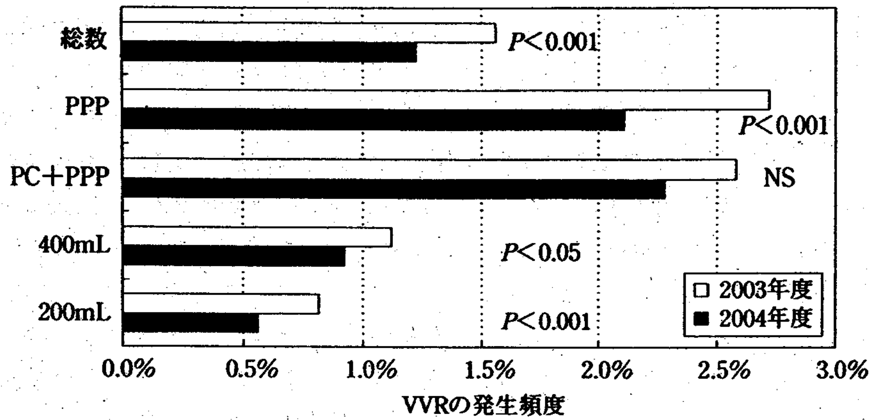

図5　埼玉赤十字血液センターにおける女性の軽症VVRの発生頻度の年度別の比較

PPP：血漿献血，PC＋PPP：血小板献血　400mL：400mLの全血献血　200mL：200mLの全血献血　NS：有意差なし

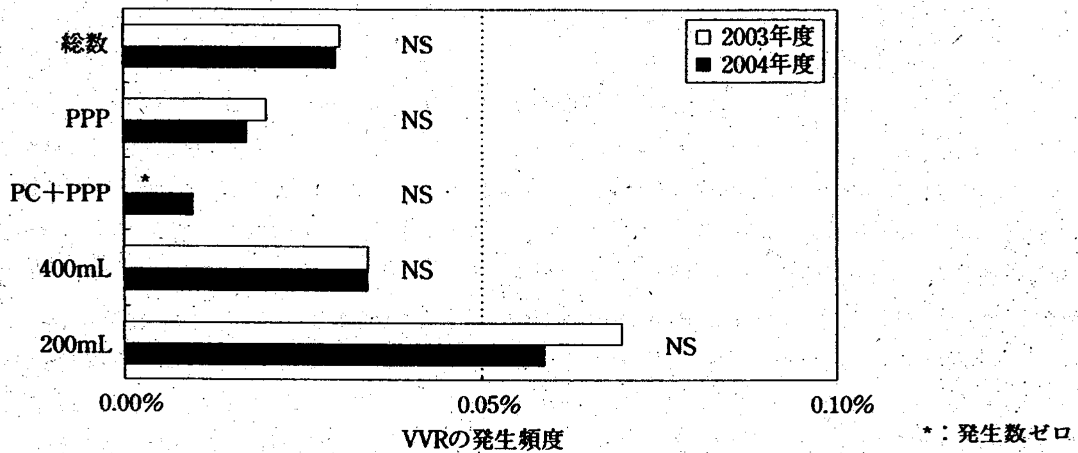

図6　埼玉赤十字血液センターにおける男性の重症VVRの発生頻度の年度別の比較

PPP：血漿献血，PC＋PPP：血小板献血　400mL：400mLの全血献血　200mL：200mLの全血献血　NS：有意差なし

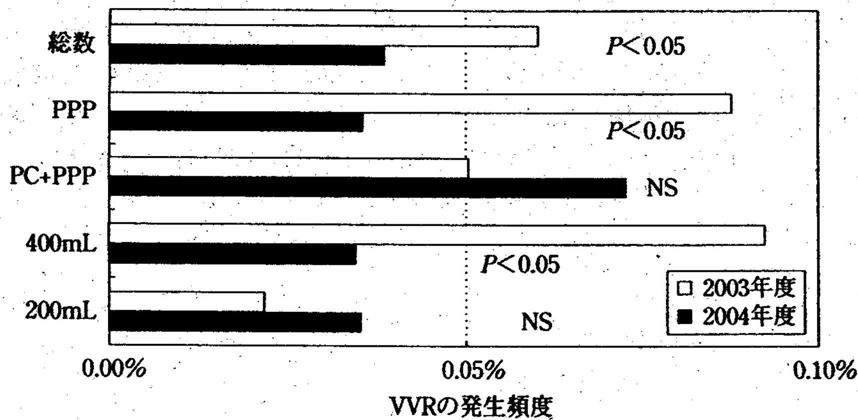

図7　埼玉赤十字血液センターにおける女性の重症VVRの発生頻度の年度別の比較

PPP：血漿献血，PC＋PPP：血小板献血　400mL：400mLの全血献血　200mL：200mLの全血献血　NS：有意差なし

献血は高校生献血を多く含むと考えられ，重症例の発生は他の献血種別より高く，パンフレットを渡す効果はみられなかった。一方，女性の重症例では，2004年度の発生率は2003年度より全体，血漿献血および400mL献血いずれも有意に低下した（図7）。しかし，200mL献血と血小板献血における重症のVVRの発生頻度はパンフレットを渡しても有意の低下はみられなかった。

考　察

今回の結果から，初回の若い全血献血の男女と中高年の成分献血の女性に少なくとも30分の休憩と水分摂取を勧めるパンフレットを渡すことは男女ともVVRの発生頻度を低下させるのに有用と考える。医療機関における医療事故の防止には患者の協力を得ることが大切とされる。今回のパンフレットを献血者に渡すことはVVR予防に献血者の協力を求めるのに役立ったのではないかと考える。またそれだけではなく，採血を担当した看護師，接遇にあたる事務職員もそのパンフレットを持つ献血者に特別な配慮をした可能性もあり，それがVVR予防に有効であった可能性がある。他のグループの献血者には少なくとも15分休むように書いた紙を渡した。このこともVVRの全体の頻度を下げるのに効果があった可能性もある。

男性で重症のVVRについてはこの方法では頻度を低下させることはできなかった。とくに，初回の若年の男性を多く含む高校生あるいは専門学校生の集団献血ではこの方法が有効でない可能性が高い。そう考える根拠は，200mL献血における重症のVVRの頻度が他の献血より高く，この男性の200mL献血はほとんどが高校生の集団献血で行われているからである。その頻度がパンフレットを渡すことで低下していないことは，これらの献血者の重症のVVRの頻度をパンフレットを渡すことでは下げることができないと考えられる。現に，10歳代の男性の初回の全血献血者に限って検索すると，データは示していないが200mL献血も400mL献血も軽症のVVRの頻度は2003年度より2004年度の方が有意に低下したが，重症のVVRはいずれの場合も有意の減少はみられなかった。したがって，初回の男性の高校生あるいは専門学校生の集団献血では重症のVVRの頻度を低下させ，さらにそれによる転倒事故を減らすためには他の方策を考える必要があると思われる。我々は10歳代と20歳代の初回の男性を多く含む高校生献血あるいは男性の専門学校生の献血では，多くの場合バスにおいて採血する。その場合に，接遇の部屋をバスから離れたところに設営するのではなく，バスのすぐそばにテントで仮の接遇の場を造り，そこに1台のバスあたり約5脚の椅子を置き，さらに専門の職員を1人配置し，椅子に座ることと水分摂取を勧め，約30分後に献血手帳を渡すようにした。そのような工夫をすることによってVVRの発生頻度は大きく変わらないが，転倒者がいなくなった。このように接遇の部屋を採血場所にできるだけ近くにすることは他の血液センターでも推奨されている[10]。今後，その効果を長期的にみていきたいと考えている。

女性の重症のVVRの頻度は血漿献血，血小板献血および400mL献血で男性より高いが，それらの頻度がパンフレットを渡すことで著しく低下した。このことは本研究が目的とした成分献血のうち血漿献血には大きな効果があったと考える。しかし，血小板献血ではその頻度が減少しなかったことは，今後の問題と思われた。200mL献血における重症例の頻度は男性より低くパンフレットを渡すようになっても有意の変化はなかった。女性の場合は，男性で200mL献血を主に行う高校生の集団献血は埼玉県では行っておらず，多くはルームなどにおける個人の献血であると思われる。したがって，そのケアも行き届いている可能性が考えられる。そのことが200mL献血において男性の重症のVVRに比し，女性の重症のVVRの頻度が低い結果に繋がった可能性がある。

VVRの減少効果がパンフレットを渡した献血者だけに限定しているか否かについて一部の献血者で検討すると，データは示していないが，10歳代の男女とも200mL献血あるいは400mL献血において初回の献血者では2003年度より2004年度の方がVVRの発生は有意に減少したが，再来の献血者では有意の減少はみられていなかった。このことはこの群ではパンフレットを渡したことがVVRの発生を低下させたと考えられる。しかし，前述のよ

うにこの群でも重症のVVRの発生には効果はなかった。また，中高年の女性の成分献血では50歳代の初回の血漿献血をした献血者のVVRだけが2003年度より2004年度の方が有意に減少していたが，50歳代の再来あるいは60歳代の初回と再来では有意の減少は認められなかった。むしろ，若年の女性の血漿献血でVVRの減少傾向がみられていた。献血者を年齢別に分けるとその群に属する献血者数やVVRを起こした献血者数が少なくなり，その効果の判定が困難になった可能性もあるが，VVR予防のためのパンフレットを渡すという行為が献血者全員と職員のVVRに対する意識を高めたことも他の群のVVRの減少に関係した可能性もあると考える。

VVRのハイリスクグループを選び，VVRに対する対策を指示するパンフレットを渡すことは，VVRの減少に一定の効果を認めた。この方法が他センターでも有効であるか否かを検証していただくことが必要ではないかと考える。さらに，全国の血液センターにおける献血時の副作用を起こした例を集め，対策をたてることとそれぞれのセンターで有効とされる対策を集めて，それらの対策を全国のセンターで実施し，その有効性を検証することが必要であろう。

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20

原　著

16，17歳（高校生）を対象とする400ml全血と成分採血導入の可否—介入試験による検討

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（平成18年4月4日受付）
（平成18年7月12日受理）

若年者（16，17歳）からの400ml全血と成分献血についての意識調査を行った．高校生（集団献血実施校，非実施校），高校教諭，父母を対象に，両採血法に関する資料（情報）を提供し，その前後で同一内容のアンケートを行った．調査対象総数は1,450人，回答数（率）は1,177人（81%）であった．前調査では，400ml全血，成分の各献血法を「可」とするのは，それぞれ67，61%，「分らない」は28，35%であったが，この「分らない」の1/3～1/2が資料提供により賛成に転じ，後調査では「可」がそれぞれ77，74%に増加した．「反対」は前後の調査とも数～10%であった．

若年者での両採血の実施については，社会的な合意は大方得られており，適切な情報の提供のもとに実施可能であると考える．

キーワード：若年献血者，400mL献血，成分献血，介入試験

はじめに

少子高齢化が進むことにより，血液の供給面では献血者層，特に若い世代の献血者数と献血率の減少[1)2)]が，需要面では高齢受血者数と受血率の増加[3)]があり，需給の不均衡を生じることが懸念される．既に両者の関連を推計した報告[4)]があるが，その後に，献血年齢の上限が69歳に引き上げられ，医療技術の進歩や適正使用の推進により新鮮凍結血漿やアルブミン製剤の供給量は明らかに減少し，MAP加赤血球濃厚液のそれは微増に留まっている[5)]ことなどにより，現在は輸血用血液の需給の均衡は維持されているが，本質的な状況に変化はないと考えられる．

このような状況から，今後の血液の量的確保対策として，16，17歳を対象に400ml全血採血と成分採血の導入の是非を検討する必要があると考え，まず社会的な合意が得られるか否かの調査を2002年に行ったところ，過半数が賛意を表したが，「分からない」との回答者が20～30%認められた[6)]．そこで，これらの採血法に関する解説資料を提供して，「分からない」との回答者がその前後でどのように意識の変化を示すのかの，介入試験を試みたので報告する．

方　法

対象者は，集団献血実施校の高校生（A群）400人，非実施校の高校生（B群）450人，およびA，B両群の教諭（C群）200人と父母（D群）400人である．調査方法は，高校生では献血に関する

Table 1　Questionnaire

Question 1.　Recently, 400 m*l* whole blood donations from young persons (high school students) aged 16 or 17 have been discussed. What do you think of this idea?

① Approve if he/she meets the criteria (body-weight etc.) defined by the Blood Collection Standards.
② Approve at or over the age of 17.
③ Approve at or over the age of 16.
④ Unclear.
⑤ Unacceptable. [Reasons：　　　　　　　　]

Question 2.　Recently, apheresis donations (collecting only platelets or plasma) from young persons (high school students) aged 16 or 17 have been discussed. What do you think of this idea?

① Approve if he/she meets the criteria (body-weight etc.) defined by the Blood Collection Standards.
② Approve at or over the age of 17.
③ Approve at or over the age of 16.
④ Unclear.
⑤ Unacceptable. [Reasons：　　　　　　　　]

アンケート調査用紙（Table 1）を配布・記入し（前調査），次いで配布した解説資料を読んでもらった後に，再度同一内容のアンケート調査用紙に記入（後調査）を依頼し，回収した．教諭と父母については，同様な手順による記入を依頼し，郵送により回収した．

解説資料の内容[7]としては，循環血液量（体重）と安全な採血量の関係，過去15年間の献血者数，採血基準の概要，400m*l*採血と成分採血の概要，前述の2002年に実施した調査結果の要約を記載した．調査期間は2003年1～2月とした．

両調査について回答の得られたものを，対象者群別に，400m*l*全血と成分採血についてクロス集計し，さらにC，D群については献血経験の有無別に，A群は献血の種類（400m*l*と200m*l*全血献血）別にも比較検討したが，B群については献血歴の有無の調査は行わなかった．なお，回答は①「体重等の基準を満たしていればやってもよい」，②「17歳以上なら可」，③「16歳以上なら可」，④「分からない」，⑤「やるべきではない」（反対）であり，①②③を賛成群として集計した．有意差検定には χ^2 検定を用いた．

成　績

1）16・17歳の400m*l*献血について

有効回答数および回答率はA，B，C，D群順に337（84%），383（85%），167（84%），290（73%），総数1,177（81%）であった．前調査と後調査の群別クロス集計をTable 2に示す．前調査での①②③の賛成回答は，A，B，C，D群順に74，55，72，70%で，B群が他群より少なく（$p<0.005$），④「わからない」は各々25，42，16，22%で，B群が他群より多く（$p<0.005$），C群はA群より少なかった（$p<0.025$）．一方，⑤「やるべきではない」は各々1，3，13，8%で，A，B群はC，D群より少なかった（$p<0.005$）．

後調査では，賛成回答がA，B，C，D群順に83，69，83，76%に増加したが，それは各群の④の32～50%および⑤の8～36%が賛成回答に移動したためである．その結果④が16，28，10，17%へと減少し，⑤もわずかながら減少した．逆に賛成回答から⑤に変わったのは，B群の0.5%とD群の1%，④へは各々4，4，1，1%と少数であった．

後調査の対象群間差をみると，賛成回答ではB群はA，C群より（$p<0.005$），D群はA群より少なく（$p<0.025$），④ではB群は他群より多くなり（$p<0.005$），⑤は変化しなかった．

即ち，資料による介入効果がみられたのは，賛成回答の増加したA，B群（$p<0.005$）とC群（$p<0.025$）であり，A，B群での④の減少であった（$p<0.005$）．

献血歴別にみると（Table 3），C群の献血歴ありは130人（78%），なしは36人，D群のありは175人（61%），なしは114人であった．C群のあり，

Table 2 Opinion and change in opinion concerning the acceptability of 400 m*l* whole blood donations from young persons before and after reading a document about 400 m*l* whole blood donations by groups.

A group		after ①	after ②	after ③	after ④	after ⑤	before total (%)
before	①	175	6	3	7	0	191 (57)
	②	6	28	1	1	0	36 (11)
	③	5	0	14	2	0	21 (6)
	④	30	6	6	43	0	85 (25)
	⑤	1	0	0	2	1	4 (1)
after total (%)		217 (64)	40 (12)	24 (7)	55 (16)	1 (0)	337

①②③ before total: 248 (74%); ①②③ after total: 281 (83%)

Change in opinion from ④ to ①②③ : 42/85 = 49%
⑤ to ①②③ : 1/4 = 25%
⑤ to ④ : 2/4 = 50%

B group		after ①	after ②	after ③	after ④	after ⑤	before total (%)
before	①	169	2	2	8	1	182 (48)
	②	7	2	0	1	0	10 (3)
	③	5	0	15	0	0	20 (5)
	④	47	10	3	97	3	160 (42)
	⑤	4	0	0	1	6	11 (3)
after total (%)		232 (61)	14 (4)	20 (5)	107 (28)	10 (3)	383

①②③ before total: 212 (55%); ①②③ after total: 266 (69%)

Change in opinion from ④ to ①②③ : 60/160 = 38%
⑤ to ①②③ : 4/11 = 36%
⑤ to ④ : 1/11 = 9%

C group		after ①	after ②	after ③	after ④	after ⑤	before total (%)
before	①	99	2	2	2	0	105 (63)
	②	2	6	0	0	0	8 (5)
	③	0	0	7	0	0	7 (4)
	④	10	1	2	12	1	26 (16)
	⑤	6	0	1	3	11	21 (13)
after total (%)		117 (70)	9 (5)	12 (7)	17 (10)	12 (7)	167

①②③ before total: 120 (72%); ①②③ after total: 138 (83%)

Change in opinion from ④ to ①②③ : 13/26 = 50%
⑤ to ①②③ : 7/21 = 33%
⑤ to ④ : 3/21 = 14%

D group		after ①	after ②	after ③	after ④	after ⑤	before total (%)
before	①	177	2	2	2	3	186 (64)
	②	3	12	0	1	0	16 (6)
	③	0	0	1	0	0	1 (0)
	④	19	1	0	39	4	63 (22)
	⑤	2	0	0	6	16	24 (8)
after total (%)		201 (69)	15 (5)	3 (1)	48 (17)	23 (8)	290

①②③ before total: 203 (70%); ①②③ after total: 219 (76%)

Change in opinion from ④ to ①②③ : 20/63 = 32%
⑤ to ①②③ : 2/24 = 8%
⑤ to ④ : 6/24 = 25%

A group : Students in high schools giving mass blood donations
B group : Students in high schools notgiving mass blood donations
C group : Teachers in these schools
D group : Parents of these students

なし, D群のあり, なしの順に前調査の賛成は各々72, 72, 70, 69%, ④は各々16, 14, 22, 21%, ⑤は同様に12, 14, 8, 10%で, 献血歴の有無による差は認められなかった. 後調査ではそれぞれが同じように④⑤から賛成へ変化し, 同様の順に賛成が84, 81, 77, 72%, ④は各々11, 6, 15, 19%となり, ⑤はC群のありとD群のなしが5, 9%になったが, C群のなしとD群のありは変化しなかった. 資料による介入効果が認められたのはC群の献血歴ありの賛成回答の増加のみ ($p<0.025$) であった.

A群の献血種別による回答を, Table 4に示す. 前調査の賛成回答は400m*l*と200m*l*献血者では各々79%, 70%で差は無かったが, 資料により400m*l*献血者の④の59%, 200m*l*献血者のそれの46%が賛成回答へと変わり, 後調査では賛成は各々90%, 80%で, 400m*l*献血者のほうが有意に多くなった ($p<0.025$). 即ち資料による介入効果は両者に認められるが400m*l*の方がより高かった ($p<0.025$, $p<0.05$).

2) 16・17歳の成分献血について

有効回答数（率）はA, B, C, D群順に, 336

Table 3　Opinion and change in opinion concerning the acceptability of 400 m*l* whole blood donations from young persons before and after reading a document about 400 m*l* whole blood donations by previous blood donations in C and D groups.

C group with previous blood donation		after ①	after ②	after ③	after ④	after ⑤	before total (%)
before	①	79	1	1	1	0	82 (63)
	②	2	3	0	0	0	5 (4)
	③	0	0	7	0	0	7 (5)
	④	8	1	0	11	1	21 (16)
	⑤	6	0	1	2	6	15 (12)
after total (%)		95 (73)	5 (4)	9 (7)	14 (11)	7 (5)	130

before ①②③: 94 (72%)
after ①②: 109 (84%)

Change in opinion from ④ to ①②③ : 9/21 = 43%
⑤ to ①②③ : 7/15 = 47%
⑤ to ④ : 2/15 = 13%

C group without blood donation		after ①	after ②	after ③	after ④	after ⑤	before total (%)
before	①	20	1	1	1	0	23 (64)
	②	0	3	0	0	0	3 (8)
	③	0	0	0	0	0	0 (0)
	④	2	0	2	1	0	5 (14)
	⑤	0	0	0	0	5	5 (14)
after total (%)		22 (61)	4 (11)	3 (8)	2 (6)	5 (14)	36

before ①②③: 26 (72%)
after ①②: 29 (81%)

Change in opinion from ④ to ①②③ : 4/5 = 80%

D group with previous blood donation		after ①	after ②	after ③	after ④	after ⑤	before total (%)
before	①	107	0	1	0	2	110 (63)
	②	2	8	0	1	0	11 (6)
	③	0	0	1	0	0	1 (1)
	④	15	0	0	22	2	39 (22)
	⑤	0	0	0	4	10	14 (8)
after total (%)		124 (71)	8 (5)	2 (1)	27 (15)	14 (8)	175

before ①②③: 122 (70%)
after ①②: 134 (77%)

Change in opinion from ④ to ①②③ : 15/39 = 38%
⑤ to ④ : 4/14 = 29%

D group without blood donation		after ①	after ②	after ③	after ④	after ⑤	before total (%)
before	①	68	2	1	2	1	74 (65)
	②	1	4	0	0	0	5 (4)
	③	0	0	0	0	0	0 (0)
	④	4	0	0	18	2	24 (21)
	⑤	2	0	0	2	7	11 (10)
after total (%)		75 (66)	6 (5)	1 (1)	22 (19)	10 (9)	114

before ①②③: 79 (69%)
after ①②: 82 (72%)

Change in opinion from ④ to ①②③ : 4/24 = 17%
⑤ to ①②③ : 2/11 = 18%
⑤ to ④ : 2/11 = 18%

C and D groups : see Table 2

Table 4　Opinion and change in opinion concerning the acceptability of 400 m*l* whole blood donations from young persons before and after reading a document about 400 m*l* whole blood donations by 400 m*l* and 200 m*l* whole blood donations at survey in A group.

400 m*l* donation		after ①②③	after ④	after ⑤	before total (%)
before	①②③	100	2	0	102 (79)
	④	16	11	0	27 (21)
	⑤	0	0	0	0 (0)
after total (%)		116 (90)	13 (10)	0 (0)	129

Change in opinion from ④ to ①②③ : 16/27 = 59%

200 m*l* donation		after ①②③	after ④	after ⑤	before total (%)
before	①②③	137	8	0	145 (70)
	④	26	31	0	57 (28)
	⑤	1	2	1	4 (2)
after total (%)		164 (80)	41 (20)	1 (0)	206

Change in opinion from ④ to ①②③ : 26/57 = 46%
⑤ to ①②③ : 1/4 = 25%
⑤ to ④ : 2/4 = 50%

A group : see Table 2

Table 5 Opinion and change in opinion concerning the acceptability of apheresis from young persons before and after reading a document about apherersis donations by groups.

A group		after ①	after ②	after ③	after ④	after ⑤	before total (%)	
before	①	163	4	0	8	0	175 (52)	230 (68%)
	②	3	26	1	3	0	33 (10)	
	③	5	1	16	0	0	22 (7)	
	④	31	8	3	64	0	106 (32)	
	⑤	0	0	0	0	0	0 (0)	
after total (%)		202 (60)	39 (12)	20 (6)	75 (22)	0 (0)	336	
		261 (78%)						

Change in opinion from ④ to ①②③ : 42/106 = 40%

B group		after ①	after ②	after ③	after ④	after ⑤	before total (%)	
before	①	162	3	1	7	0	173 (45)	198 (51%)
	②	4	4	0	1	0	9 (2)	
	③	5	0	11	0	0	16 (4)	
	④	62	9	4	103	2	180 (47)	
	⑤	0	0	0	2	5	7 (2)	
after total (%)		233 (61)	16 (4)	16 (4)	113 (29)	7 (2)	385	
		265 (69%)						

Change in opinion from ④ to ①②③ : 75/180 = 42%
⑤ to ④ : 2/7 = 29%

C group		after ①	after ②	after ③	after ④	after ⑤	before total (%)	
before	①	92	2	1	3	0	98 (59)	109 (66%)
	②	1	3	0	0	0	4 (2)	
	③	0	0	7	0	0	7 (4)	
	④	19	1	3	19	1	43 (26)	
	⑤	3	0	1	1	8	13 (8)	
after total (%)		115 (70)	6 (4)	12 (7)	23 (14)	9 (5)	165	
		133 (81%)						

Change in opinion from ④ to ①②③ : 23/43 = 53%
⑤ to ①②③ : 4/13 = 31%
⑤ to ④ : 1/13 = 8%

D group		after ①	after ②	after ③	after ④	after ⑤	before total (%)	
before	①	156	1	1	4	1	163 (56)	184 (63%)
	②	8	12	0	0	0	20 (7)	
	③	0	0	1	0	0	1 (0)	
	④	32	1	0	53	3	89 (30)	
	⑤	1	0	0	3	15	19 (7)	
after total (%)		197 (67)	14 (5)	2 (1)	60 (21)	19 (7)	292	
		213 (73%)						

Change in opinion from ④ to ①②③ : 33/89 = 37%
⑤ to ①②③ : 1/19 = 5%
⑤ to ④ : 3/19 = 16%

A, B, C and D groups : see Table 2.

(84%), 385 (86%), 165 (83%), 292 (73%) で, 総数 1,178 (81%) であり, Table 5 に前調査と後調査の群別クロス集計を示す. 前調査では, A, B, C, D 群順に賛成が 68, 51, 66, 63% で, 400 ml 献血に対する賛成回答より 4~7% 少なかったが, 同様の傾向であり, B 群では他群より少なかった ($p<0.005$). ④「わからない」は各々 32, 47, 26, 30% で, B 群が他群より多かった ($p<0.005$). ⑤「やるべきではない」は 0, 2, 8, 7% と少数であり, A, B 群は C, D 群より少なかった ($p<0.005$).

資料読後には, ④では各群とも 37~53% が, ⑤では A, B 群は変化なく C, D 群で各々の 31, 5% が賛成回答に変ったことから, 後調査での賛成は A, B, C, D 群順に 78, 69, 81, 73% に増加し, ④は各々 22, 29, 14, 21% に減少し, C 群では⑤もわずかながら減少した. 賛成回答から⑤にかわったのは D 群の 0.5% のみ, ④へは各々 5, 4, 3, 2% であった. その結果, 後調査の対象群間差は, 賛成回答では B 群は A, C 群 ($p<0.01$, 0.005) より少なく, ④では B 群は他群より ($p<0.005$~0.05), A 群は C 群より ($p<0.05$) 多かった. ⑤では C, D 間以外はすべての群間に差を認めた ($p<0.005$~0.025).

即ち, 資料による介入効果はすべての群にみられ, 賛成回答は有意に増加 (A 群 ($p<0.01$), B, C 群 ($p<0.005$), D 群 ($p<0.025$)) し, ④は有意に減少 (A, C, D 群 ($p<0.01$), B 群 ($p<0.005$)) した.

Table 6 Opinion and change in opinion concerning the acceptability of apheresis from young persons before and after reading a document about apherersis donations by previous blood donations in C and D groups.

C group with previous blood donation		after					before total (%)	
		①	②	③	④	⑤		
before	①	73	1	1	2	0	77 (59)	86 (66%)
	②	1	2	0	0	0	3 (2)	
	③	0	0	6	0	0	6 (5)	
	④	17	1	0	15	1	34 (26)	
	⑤	3	0	1	1	5	10 (8)	
after total (%)		94 (72)	4 (3)	8 (6)	18 (14)	6 (5)	130	
		106 (82%)						

Change in opinion from ④ to ①②③ : 18/34 = 53%
⑤ to ①②③ : 4/10 = 40%
⑤ to ④ : 1/10 = 10%

C group without blood donation		after					before total (%)	
		①	②	③	④	⑤		
before	①	20	1	0	1	0	22 (61)	24 (67%)
	②	0	1	0	0	0	1 (3)	
	③	0	0	1	0	0	1 (3)	
	④	2	0	2	5	0	9 (25)	
	⑤	0	0	0	0	3	3 (8)	
after total (%)		22 (61)	2 (6)	3 (8)	6 (17)	3 (8)	36	
		27 (75%)						

Change in opinion from ④ to ①②③ : 4/9 = 44%

D group with previous blood donation		after					before total (%)	
		①	②	③	④	⑤		
before	①	92	0	1	1	1	95 (54)	108 (62%)
	②	6	6	0	0	0	12 (7)	
	③	0	0	1	0	0	1 (1)	
	④	23	1	0	31	2	57 (33)	
	⑤	0	0	0	3	7	10 (6)	
after total (%)		121 (69)	7 (4)	2 (1)	35 (20)	10 (6)	175	
		130 (74%)						

Change in opinion from ④ to ①②③ : 24/57 = 42%
⑤ to ④ : 3/10 = 30%

D group without blood donation		after					before total (%)	
		①	②	③	④	⑤		
before	①	63	1	0	3	0	67 (59)	75 (66%)
	②	2	6	0	0	0	8 (7)	
	③	0	0	0	0	0	0 (0)	
	④	9	0	0	20	1	30 (26)	
	⑤	1	0	0	0	8	9 (8)	
after total (%)		75 (66)	7 (6)	0 (0)	23 (20)	9 (8)	114	
		82 (72%)						

Change in opinion from ④ to ①②③ : 9/30 = 30%
⑤ to ①②③ : 1/9 = 11%

C and D groups : see Table 2

献血歴別にみると（Table 6），C群の献血歴あり，なし，D群の献血歴あり，なし順に前調査の賛成は各々66，67，62，66%，④は各々26，25，33，26%，⑤は各々8，8，6，8%で，献血歴の有無による差は認められなかった．後調査では，賛成が各々82，75，74，72%，④は各々14，17，20，20%，⑤はC群献血歴ありのみ減少して5%になったが，後調査でも献血歴による差は認められなかった．一方，資料による介入効果が有意に認められたのは，C，D群ともに献血歴ありのみで，両群の賛成の増加（$p<0.005$，0.025）と④の減少（$p<0.025$，0.01）およびC群の⑤の減少（$p<0.05$）であった．

A群の献血種別による回答を，Table 7に示す．前調査の賛成率は400m*l*献血者では77%と200m*l*献血者の64%より多く（$p<0.025$），後調査では，400m*l*献血者の④の57%，200m*l*献血者の33%が賛成に変わったことから，後調査の賛成は各々88%と72%になった（$p<0.005$）が，介入効果が有意であったのは400m*l*献血者のみであった（$p<0.025$）．

3）反対意見の理由

⑤「やるべきではない」との回答の理由については，400m*l*，成分献血の導入に共通しており，C群では未だ成長過程にある，体力面での不安がある，大人（18歳あるいは20歳）になってからでよい，最近の高校生は弱くなっている，等が挙げられていた．またD群ではC群と同様の理由の他

Table 7 Opinion and change in opinion concerning the acceptability of apheresis from young persons before and after reading a document about apheresis donations by 400 ml and 200 ml whole blood donations at survey in A group.

400 ml donation		after			before total (%)
		①②③	④	⑤	
before	①②③	96	3	0	99 (77)
	④	17	13	0	30 (23)
	⑤	0	0	0	0 (0)
after total (%)		113 (88)	16 (12)	0 (0)	129

Change in opinion from ④ to ①②③ : 17/30 = 57%

200 ml donation		after			before total (%)
		①②③	④	⑤	
before	①②③	123	8	0	131 (64)
	④	25	50	0	75 (36)
	⑤	0	0	0	0 (0)
after total (%)		148 (72)	58 (28)	0 (0)	206

Change in opinion from ④ to ①②③ : 25/75 = 33%

A group : see Table 2

に，本人に正しい判断が望めない，成分採血時の感染が恐い，フィルター経由の環流（返血）は不可，との回答があった．これらの見解は資料を読んだ後でもほとんどの回答で変化はなく，献血経験の有無による差も認められなかったが，保護者の許可を条件とするとの⑤から④への変更が，C群に1人あった．

前調査の賛成回答から⑤への変更では，B群で量が多い，D群で正しい判断が望めない，他の方法を考えるべきとの理由が挙げられていたが，④への変更には理由の記載はなかった．

考　察

今後予測される血液不足対策としては，献血量の増量と使用適正化による量的抑制とが必要である．前者については，1986年の400ml全血採血と成分採血の導入，1999年の年齢の上限の69歳への引き上げとがあり，いずれも量的確保に効果的であった．今後の献血量の確保対策としては，先ずは現行の採血基準に該当する年齢層のより多くの参加を求める努力をすることであるが，さらには現在200mlの全血献血しかできない16，17歳の若年者（高校生）を対象にして，400ml全血と成分献血を導入することの是非を検討することである．

近年の年齢階級別の人口に対する献血率の推移をみると，毎年若年者ほど高い傾向にあるが，16～19歳の献血率は1985年をピークに以降の低下傾向が顕著である[1)2)]．このような低下傾向の理由の一つとして，医療機関の血液使用状況が200ml全血由来から400ml全血由来へと大幅に移行し，200ml全血由来の赤血球成分の使用量が激減してきていることから，日赤血液センターでは200ml全血採血を抑制する方針であることも挙げられる．しかしながら，献血のきっかけとして高校生献血を挙げる献血者が多いとの報告があり[6)]，高校生献血がその後の献血指向性に大きな役割を持っているといえることから，より合理的な高校生献血を推進することが必要と考えられる．

採血基準は，医学的な安全性とともに，社会的な合意が得られなければならない．1986年の採血基準改訂時には，400ml全血採血と成分採血時の安全性を循環血液量に対する採血量の比として検討し，それが12～13％以内（体重約50kgで400ml採血）であれば問題はないとされ[8)]，同様なことは他にも報告されている[9)]．このことは年齢には関係しないと考えられ，事実自己血輸血では16歳未満あるいは70歳以上でも採血が行われているが，特に年齢による問題点は指摘されていない．しかし，1986年の採血基準の制定時には社会的に受入れ易いことを考慮して，18歳以上とされた経緯がある．

今回のアンケート調査では，400ml全血献血で67％，成分献血で61％が，主に体重等の採血基準を満たしていれば16，17歳での導入に賛成していることから，現在では大方の合意は得られているものと考えられる．このことは，両採血法への理解が導入後20年近く大過なく行われてきていることから，より深まってきていることの表れと

もいえるであろう．さらに，前調査で400ml全血献血について「分らない」と回答した中の32～50%が，B群（献血非実施校）を含めて資料提供後に賛成に転じたこと，さらに成分献血についても同様に「分らない」との回答中の37～53%が賛成に変わったこと，しかも「やるべきではない」(反対者）の人数は少ないものの資料提供後には不変ないしわずかな減少であったことは，400ml全血や成分献血についての実態を理解することにより，賛成者が増加することを示している．また，C，D群（教諭，父母）では献血経験者の方が，またA群（献血実施校）では200ml献血者より400ml献血者のほうが，資料提供後の賛成への転換率が高かった．高校生の多くは初回は200ml献血であることも考慮すれば，献血経験が資料内容の理解をより容易にする効果があると考えられる．

海外での状況としては，欧米での採血基準（主に採血量と年齢）を各国のホームページ等で検索した結果，全血採血は体重50kg以上，採血量450～500mlの場合，年齢の下限は17あるいは18歳が多かったが，米国では一般には17歳[10]としているものの，ニューヨーク，カリフォルニア等の7州では16歳でも親の同意があればよく，またオーストラリアでも16，17歳の採血には親の同意を必要としている．なお，ニューヨーク州が16歳からとしたのは2005年4月であり[11]，今後はその他の州においても年齢の下限の見直しが行われるものと思われる．

以上のごとく，今回のアンケート調査結果や国外の状況からして，16，17歳での400ml全血および成分採血の実施は可能であると考える．本邦ではすでに200ml全血採血が16歳から行われている状況を踏まえれば，親権者の同意の必要性については今後検討すべき課題であろう．

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INTRODUCTION OF 400 ML WHOLE BLOOD AND APHERESIS DONATIONS FROM AGE 16 AND 17 (HIGH SCHOOL STUDENTS) INTO THE BLOOD PROGRAM —INVESTIGATION OF CHANGING OPINIONS BEFORE AND AFTER REVIEW OF EXPLANATORY DOCUMENTS—

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In order to obtain more blood for an increasingly aged society, a questionnaire survey was conducted to discover whether it would be socially acceptable to accept 400 m*l* whole blood (WB) and apheresis donations for the blood program from young persons of the age of 16 and 17 (mainly high school students), who are presently permitted to donate 200 m*l* WB only. We surveyed high school students who did and did not participate in mass blood donations in schools, their high school teachers, and parents. They were asked to reply to the same questions before and after reading documents explaining both blood donation types. The total number of respondents (rate) was 1,450 (81%). Before reviewing the documents 67% answered "acceptable" to 400 m*l* WB and 61% to apheresis, and 28% and 35% answered "unclear", respectively. One-third to one-half of those who answewrd "unclear" changed their opinion to "acceptable" after reading the documents. This resulted in an increase of "acceptable" opinions to 77% for 400 m*l* WB and to 74% for apheresis. The proposal was "declined" by around 10% or less in both questions.

It is considered that the introduction of 400 m*l* WB and apheresis donations from young persons into the blood program would be commonly accepted after informed consent was obtained, and that the provision of suitable information on these donations can gain lead to an increase in acceptability.

Key words: Young donors, 400 m*l* donation, apheresis donation, intervention survey

資料2－3②

採血基準に関する各種論文

（第2回採血基準見直しの検討に係るﾜｰｷﾝｸﾞｸﾞﾙｰﾌﾟ追加提示分）

平成19年度 10代年齢別採血副作用発生率

VVR発生件数

採血種類	性別	16歳	17歳	18歳	19歳
200mL	男	223	270	119	76
	女	304	353	354	283
	男女	527	623	473	359
400mL	男	−	−	1,347	1,333
	女	−	−	702	703
	男女	−	−	2,049	2,036
PPP	男	−	−	32	30
	女	−	−	224	297
	男女	−	−	256	327
PC+PPP	男	−	−	39	74
	女	−	−	156	244
	男女	−	−	195	318

VVR発生率

採血種類	性別	16歳	17歳	18歳	19歳
200mL	男	1.370%	1.155%	2.039%	3.196%
	女	1.714%	1.456%	1.872%	1.725%
	男女	1.549%	1.308%	1.912%	1.911%
400mL	男	−	−	2.673%	2.347%
	女	−	−	3.460%	2.985%
	男女	−	−	2.899%	2.534%
PPP	男	−	−	1.454%	1.008%
	女	−	−	3.484%	2.891%
	男女	−	−	2.966%	2.468%
PC+PPP	男	−	−	0.989%	1.043%
	女	−	−	4.544%	3.853%
	男女	−	−	2.644%	2.368%

VVR重症発生件数

採血種類	性別	16歳	17歳	18歳	19歳	
200mL	男	6	5	3	1	15
	女	10	14	8	3	35
	男女	16	19	11	4	50
400mL	男	−	−	51	41	92
	女	−	−	32	31	63
	男女	−	−	83	72	##
PPP	男	−	−	0	0	0
	女	−	−	7	9	16
	男女	−	−	7	9	16
PC+PPP	男	−	−	0	1	1
	女	−	−	5	13	18
	男女	−	−	5	14	19

VVR重症発生率

採血種類	性別	16歳	17歳	18歳	19歳
200mL	男	0.037%	0.021%	0.051%	0.042%
	女	0.056%	0.058%	0.042%	0.018%
	男女	0.047%	0.040%	0.044%	0.021%
400mL	男	−	−	0.101%	0.072%
	女	−	−	0.158%	0.132%
	男女	−	−	0.117%	0.090%
PPP	男	−	−	0.000%	0.000%
	女	−	−	0.109%	0.088%
	男女	−	−	0.081%	0.068%
PC+PPP	男	−	−	0.000%	0.014%
	女	−	−	0.146%	0.205%
	男女	−	−	0.068%	0.104%

VVR転倒発生件数

採血種類	性別	16歳	17歳	18歳	19歳
200mL	男	6	4	0	0
	女	2	6	6	6
	男女	8	10	6	6
400mL	男	−	−	21	24
	女	−	−	20	18
	男女	−	−	41	42
PPP	男	−	−	0	0
	女	−	−	2	1
	男女	−	−	2	1
PC+PPP	男	−	−	1	1
	女	−	−	3	2
	男女	−	−	4	3

VVR転倒発生率

採血種類	性別	16歳	17歳	18歳	19歳
200mL	男	0.037%	0.017%	0.000%	0.000%
	女	0.011%	0.025%	0.032%	0.037%
	男女	0.024%	0.021%	0.024%	0.032%
400mL	男	−	−	0.042%	0.042%
	女	−	−	0.099%	0.076%
	男女	−	−	0.058%	0.052%
PPP	男	−	−	0.000%	0.000%
	女	−	−	0.031%	0.010%
	男女	−	−	0.023%	0.008%
PC+PPP	男	−	−	0.025%	0.014%
	女	−	−	0.087%	0.032%
	男女	−	−	0.054%	0.022%

皮下出血発生件数

採血種類	性別	16歳	17歳	18歳	19歳
200mL	男	7	10	5	1
	女	46	54	32	32
	男女	53	64	37	33
400mL	男	−	−	33	49
	女	−	−	32	47
	男女	−	−	65	96
PPP	男	−	−	12	17
	女	−	−	82	115
	男女	−	−	94	132
PC+PPP	男	−	−	24	50
	女	−	−	39	84
	男女	−	−	63	134

皮下出血発生率

採血種類	性別	16歳	17歳	18歳	19歳
200mL	男	0.043%	0.043%	0.086%	0.042%
	女	0.259%	0.223%	0.169%	0.195%
	男女	0.156%	0.134%	0.150%	0.176%
400mL	男	−	−	0.065%	0.086%
	女	−	−	0.158%	0.200%
	男女	−	−	0.092%	0.119%
PPP	男	−	−	0.545%	0.571%
	女	−	−	1.275%	1.119%
	男女	−	−	1.089%	0.996%
PC+PPP	男	−	−	0.609%	0.705%
	女	−	−	1.136%	1.326%
	男女	−	−	0.854%	0.998%

2009/2/2 採血基準見直しの検討に係るワーキンググループ

穿刺部痛発生件数

採血種類	性別	16歳	17歳	18歳	19歳
200mL	男	3	5	2	1
	女	4	8	9	7
	男女	7	13	11	8
400mL	男	−	−	10	7
	女	−	−	6	5
	男女	−	−	16	12
PPP	男	−	−	2	0
	女	−	−	2	9
	男女	−	−	4	9
PC+PPP	男	−	−	0	5
	女	−	−	5	6
	男女	−	−	5	11

穿刺部痛発生率

採血種類	性別	16歳	17歳	18歳	19歳
200mL	男	0.018%	0.021%	0.034%	0.042%
	女	0.023%	0.033%	0.048%	0.043%
	男女	0.021%	0.027%	0.044%	0.043%
400mL	男	−	−	0.020%	0.012%
	女	−	−	0.030%	0.021%
	男女	−	−	0.023%	0.015%
PPP	男	−	−	0.091%	0.000%
	女	−	−	0.031%	0.088%
	男女	−	−	0.046%	0.068%
PC+PPP	男	−	−	0.000%	0.070%
	女	−	−	0.146%	0.095%
	男女	−	−	0.068%	0.082%

平成19年度:献血者数

採血種類	性別	16歳	17歳	18歳	19歳
200mL	男	16,277	23,376	5,836	2,378
	女	17,736	24,248	18,908	16,404
	男女	34,013	47,624	24,744	18,782
400mL	男	−	−	50,386	56,791
	女	−	−	20,288	23,548
	男女	−	−	70,674	80,339
PPP	男	−	−	2,201	2,976
	女	−	−	6,430	10,273
	男女	−	−	8,631	13,249
PC+PPP	男	−	−	3,943	7,094
	女	−	−	3,433	6,333
	男女	−	−	7,376	13,427

17歳男性の400mL全血採血に関する検討

②

【方法】

＜供血者の選択＞

1）採血時の満年齢が17歳であること

2）現行の400ml全血採血の基準を満たすこと

3）文書により本人および親権者の同意がえられること

4）各施設50名（北海道、宮城県、東京都、愛知県、大阪府、岡山県、福岡県）

＜検討項目＞

1)採取中・採取後の副作用の有無と採血後1週間以内の自覚症状の有無（アンケート調査）

2）赤血球採取前後の供血者の検査項目

採血前，採血3か月後に血球計数，血清鉄，TIBC，フェリチン値について検査

＜コントロール群＞

1）現行採血基準で400ml全血採血を行なっている18歳・19歳の献血者

2）各施設50名（北海道、宮城県、東京都、愛知県、大阪府、岡山県、福岡県）

*本研究は、試験プロトコール等について東京医科歯科大学の医学倫理委員会の承認を得ている

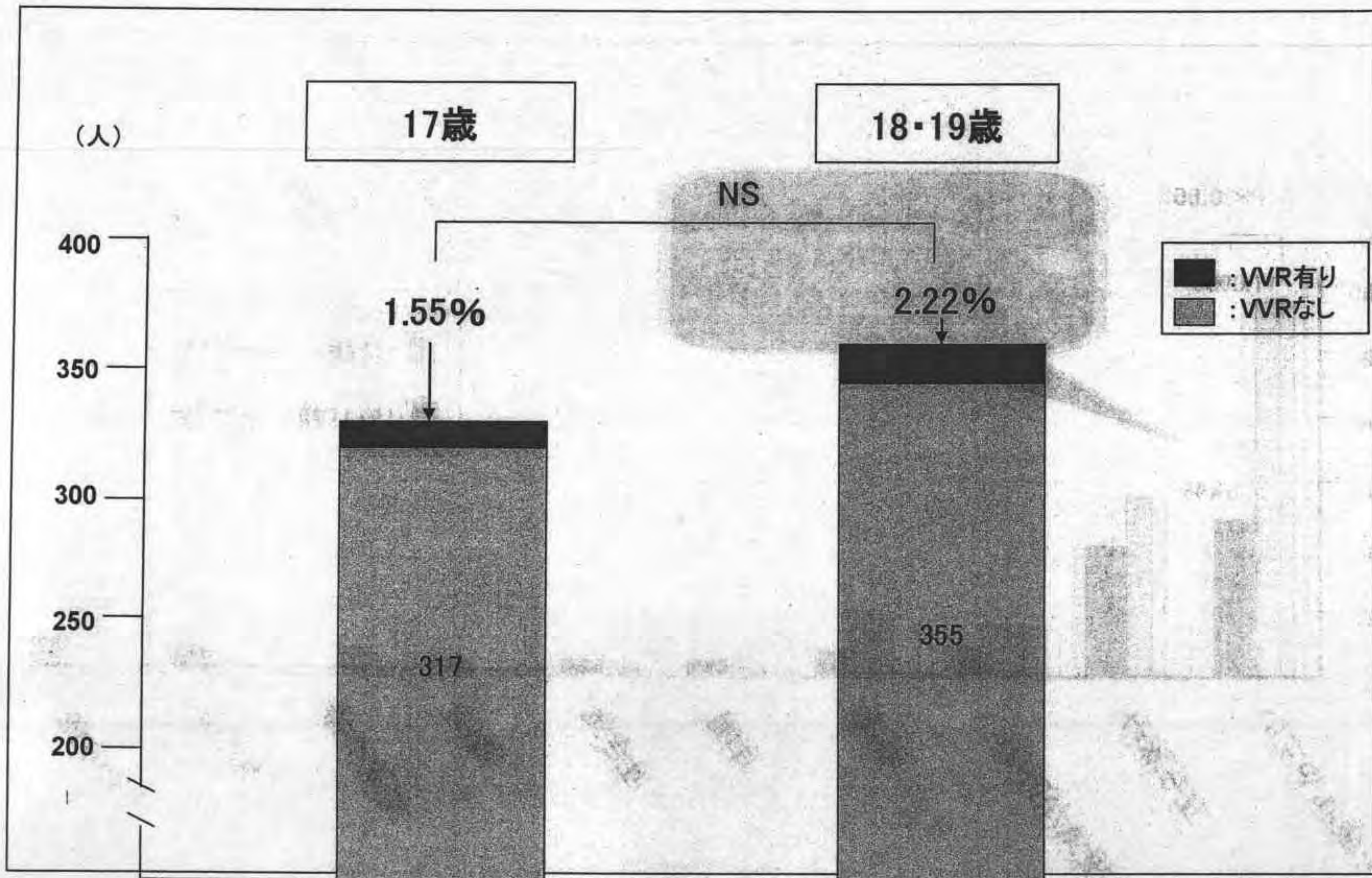
表1. 施設別400ml 採血例数

	17歳男性(検討群)	18・19歳男性(コントロール群)
北海道センター	45	46
宮城センター	43	57
東京都センター	65	58
愛知センター	43	45
大阪センター	53	57
岡山センター	21	45
福岡センター	52	55
計	322	363

【結果】　Ⅰ.供血者の背景

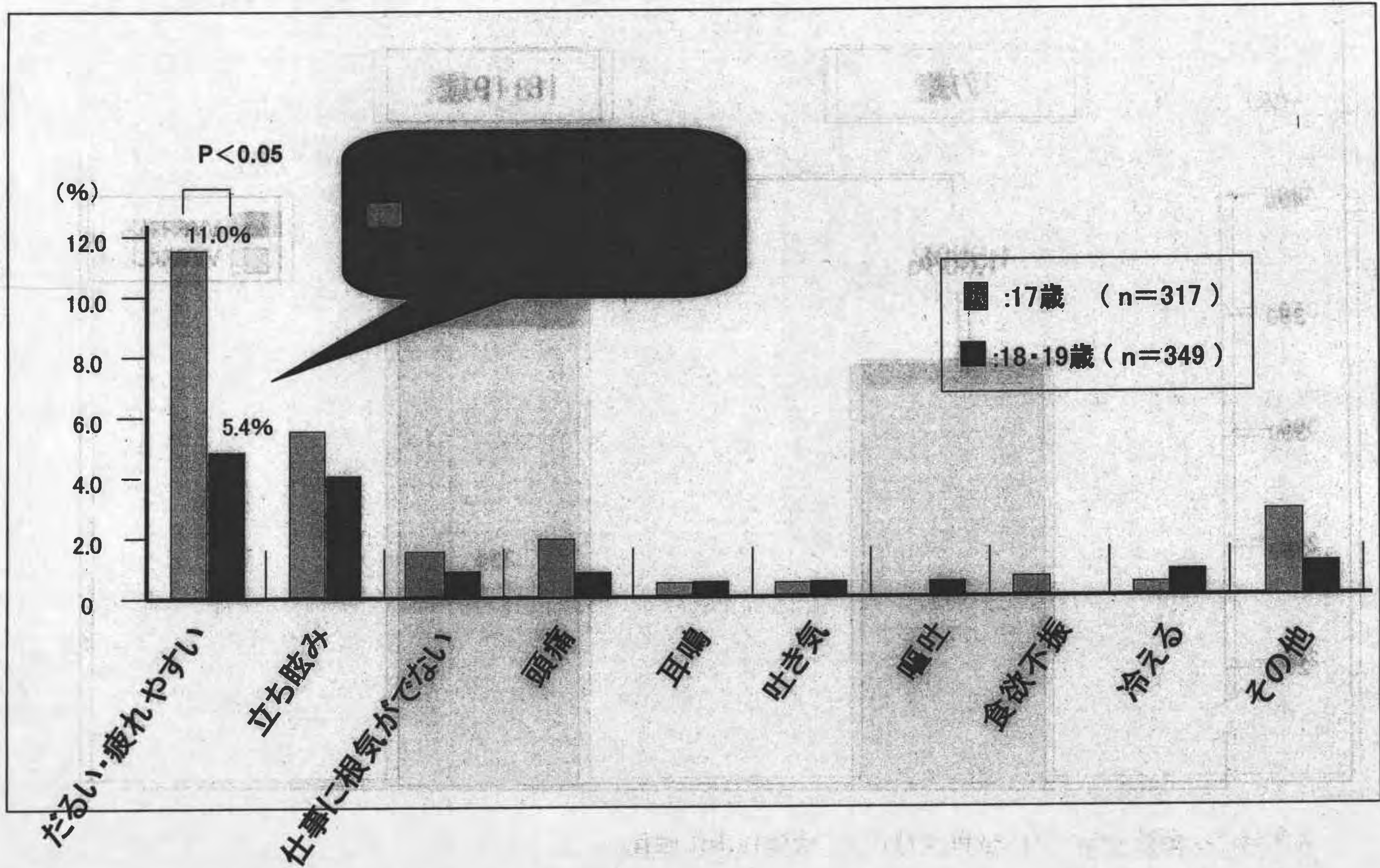
対象		17歳(検討群)	18・19歳(コントロール群)	有意差
例数		322	363	
年齢	(歳)	17.6 ± 0.3	19.0 ± 0.5	
身長	(cm)	171.1 ± 5.4	171.7 ± 5.6	NS
体重	(kg)	64.8 ± 10.2 (50 - 112)	64.6 ± 8.7 (51 – 98)	NS
循環血液量	(ml)	4526 ± 549 (3672 – 7074)	4529 ± 467 (3620 – 6276)	NS
採血量	(ml)	398.7 ± 19.2	399.2 ±13.9	NS
採血量/循環血液量	(%)	8.9 ± 1.0	8.9 ± 0.9	NS

有意差検定:Student t-test （$p<0.05$）

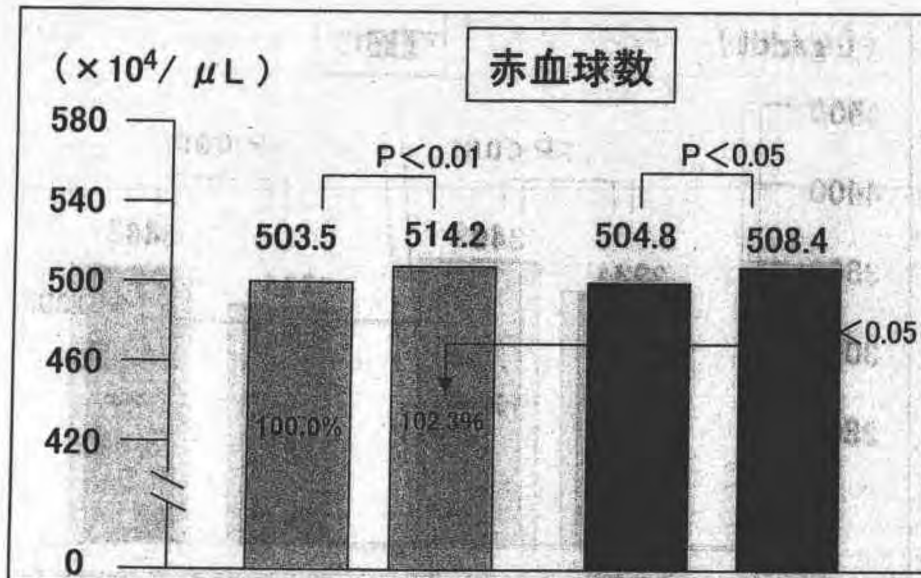


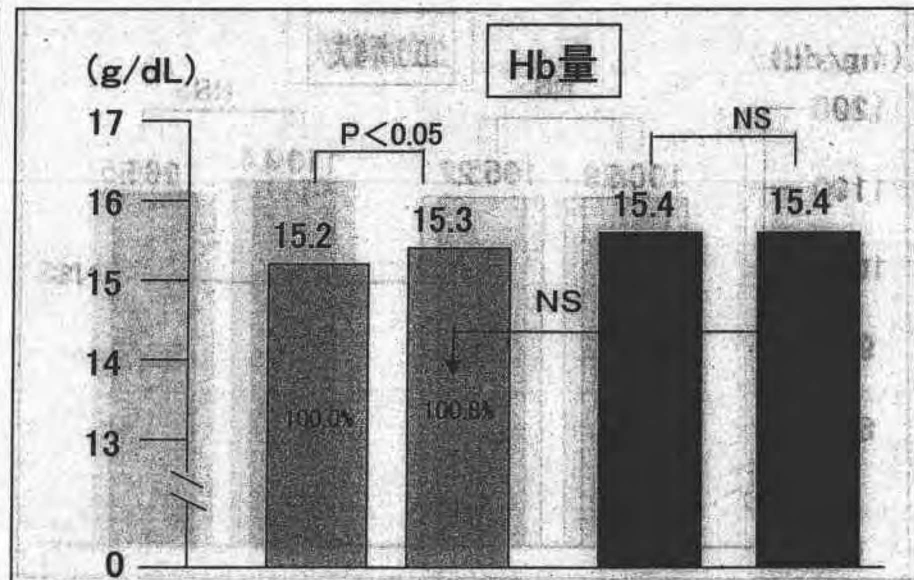
*全症例とも投薬することなく仰臥安静にて1時間以内に回復

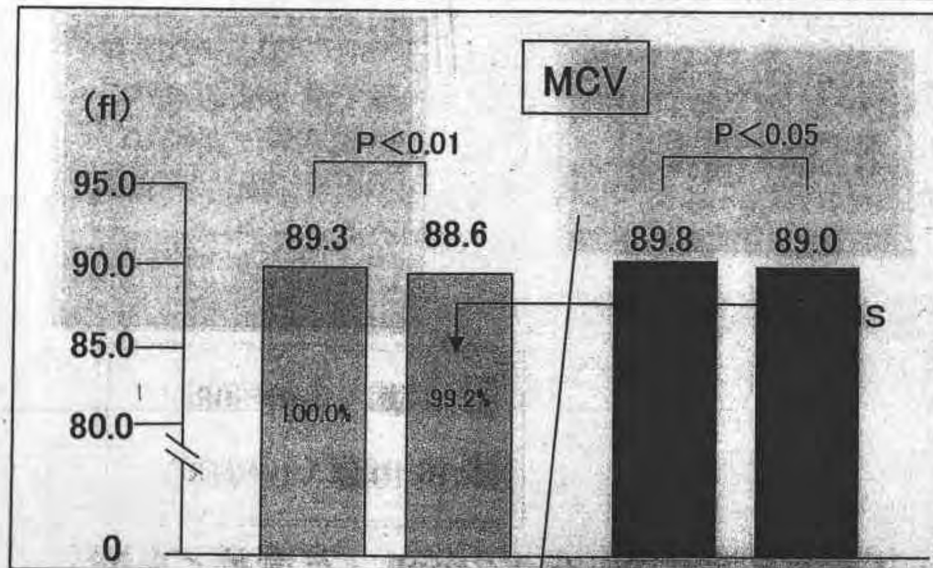
**有意差検定:2x2 Chi square test and Fisher's test ($p<0.05$)



*有意差検定:2x2 Chi square test and Fisher's test （p＜0.05）



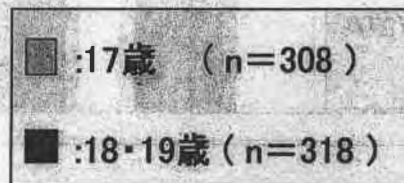




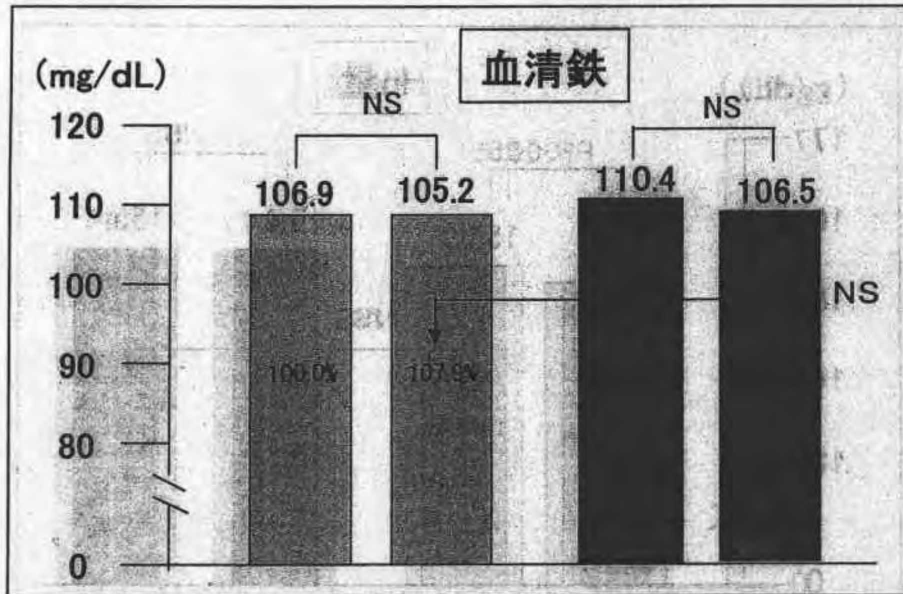
3か月後のHb値が献血基準の12.5g/dLに満たなかった例

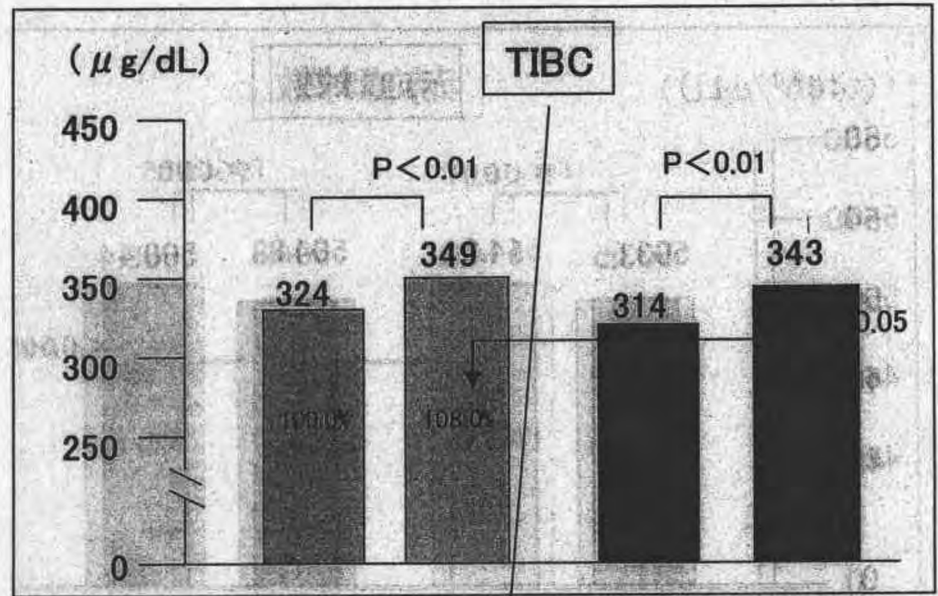
17歳：　1例(0.3%)：採血前Hb値(12.6g/dL)

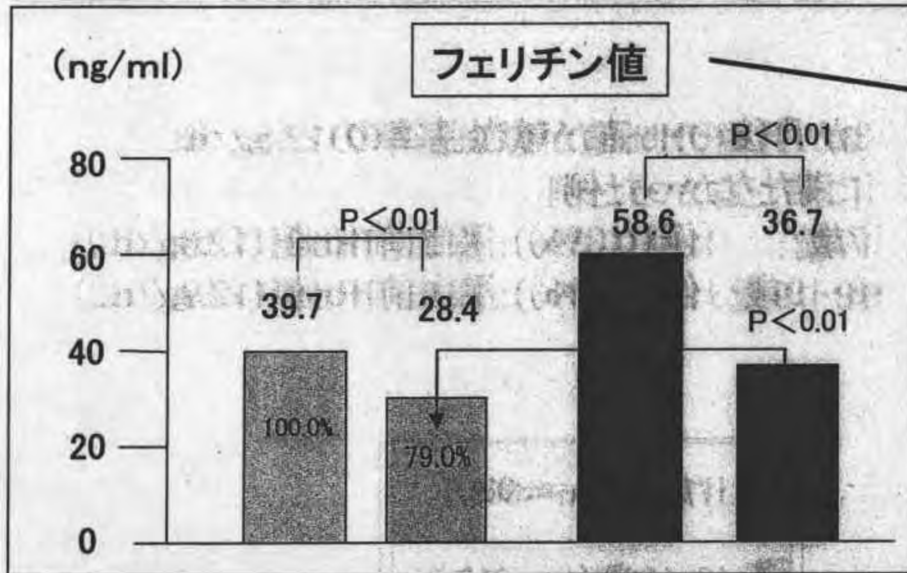
18・19歳：1例(0.3%)：採血前Hb値(12.9g/dL)



*有意差検定:Student t-test ($p<0.05$)

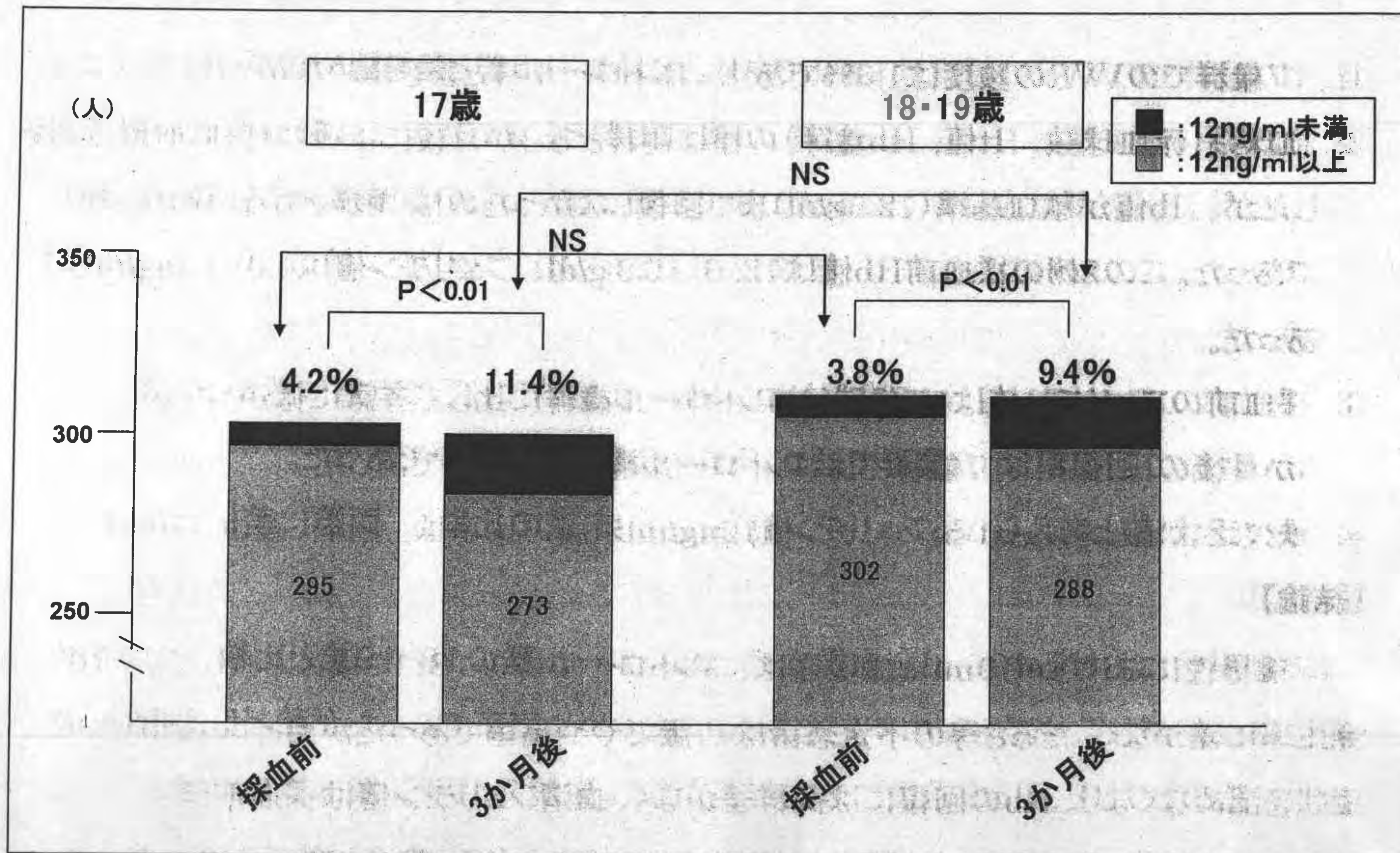






:17歳　(n＝308)
:18・19歳 (n＝318)

*有意差検定:Student t-test （有意差＜0.05）



*有意差検定:2x2 Chi square test and Fisher's test （p＜0.05）

17歳男性の400ml全血採血(まとめ)

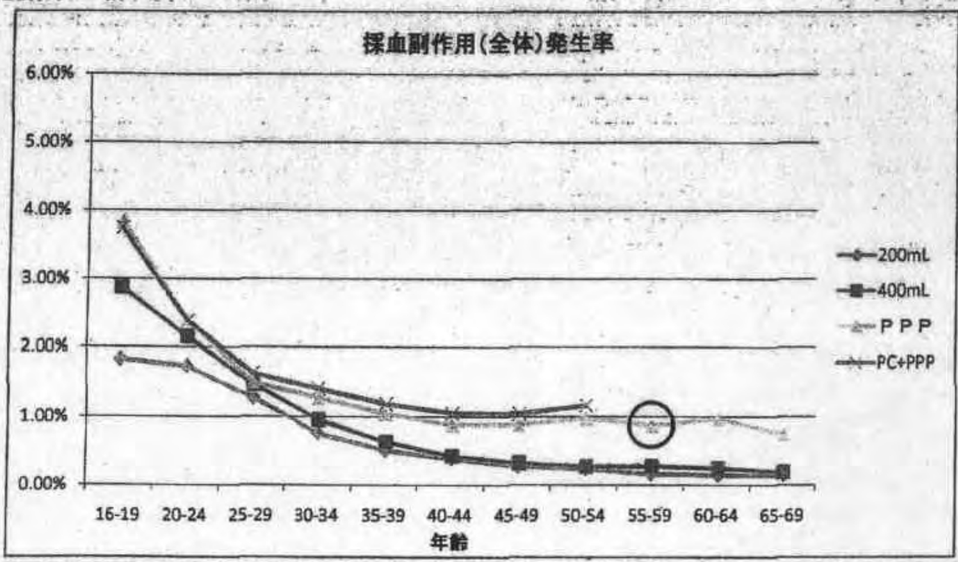
1. 17歳群でのVVRの頻度は1.6%であり、コントロール群と差を認めなかった。
2. 血球系(赤血球数, Ht値, Hb量等)の値は両群とも3か月後には概ね採血前値に回復したが、Hb値が献血基準(12.5g/dl)まで回復しなかったのは 両群とも各1例(0.3%)であった。この2例の採血前Hb値は12. 6, 12.9 g/dl、フェリチン値は4.0, 6.3ng/ml であった。
3. 採血前のフェリチン値は17歳群はコントロール歳群に比して有意に低かったが、3か月後の回復率は17歳群ではコントロール群より速やかであった。
4. 鉄欠乏状態と考えられるフェリチン値12ng/ml未満の比率は, 両群に差はなかった。

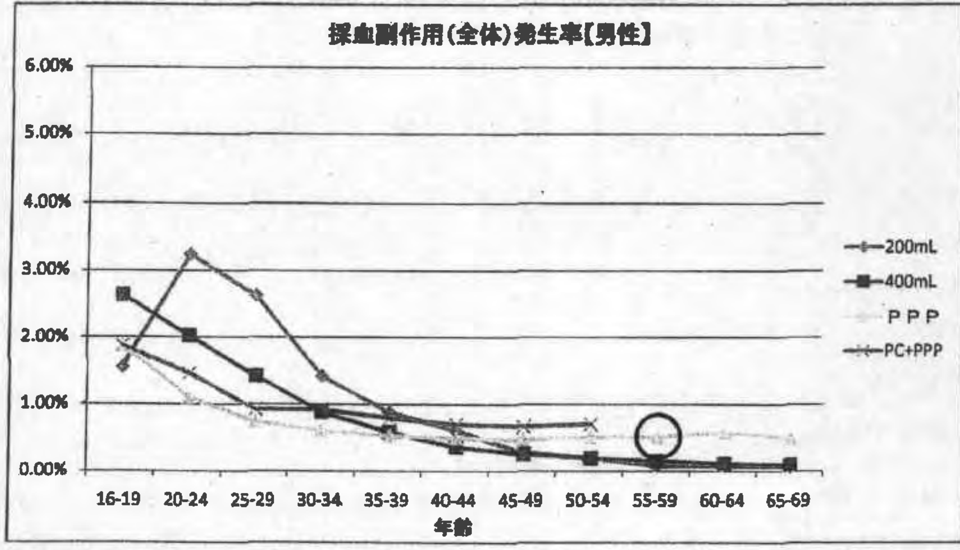
【結論】

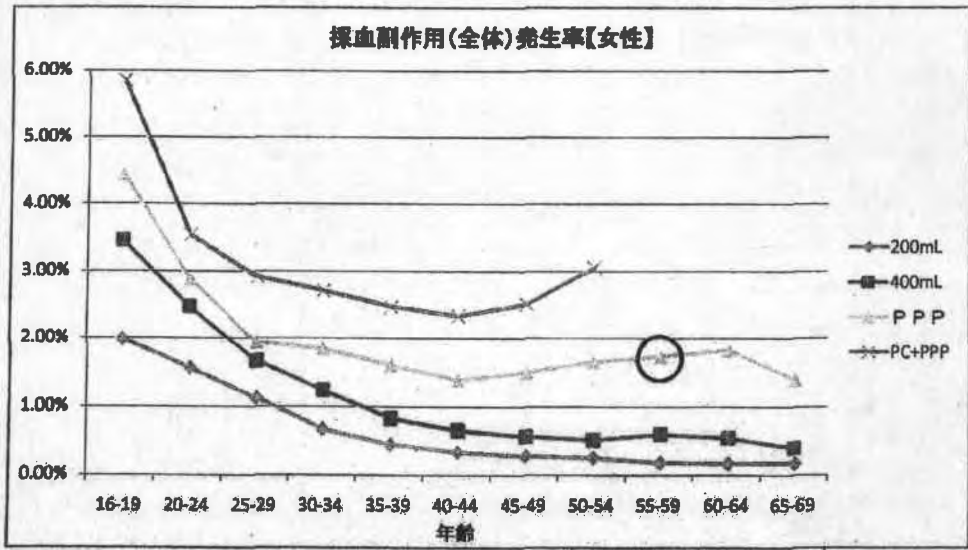
17歳男性における400ml全血採血は、コントロール群の18・19歳と比較してVVRの発生率に差がなく、だるさ等の不定愁訴は17歳でやや高率であったが殆どは1週間以内に症状を認めなくなり、Hbの回復には両群差がなく、血清フェリチン値は採血前値でやや低い傾向は認めたが、回復はより速やかであることから、安全に施行可能と考える。

③

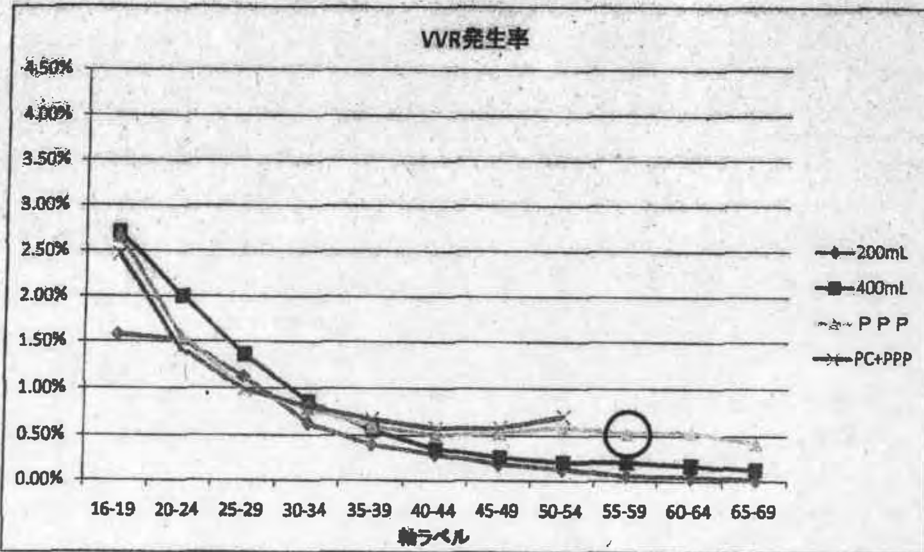
採血副作用発生率（年齢別・性別・採血種類別：平成19年度）

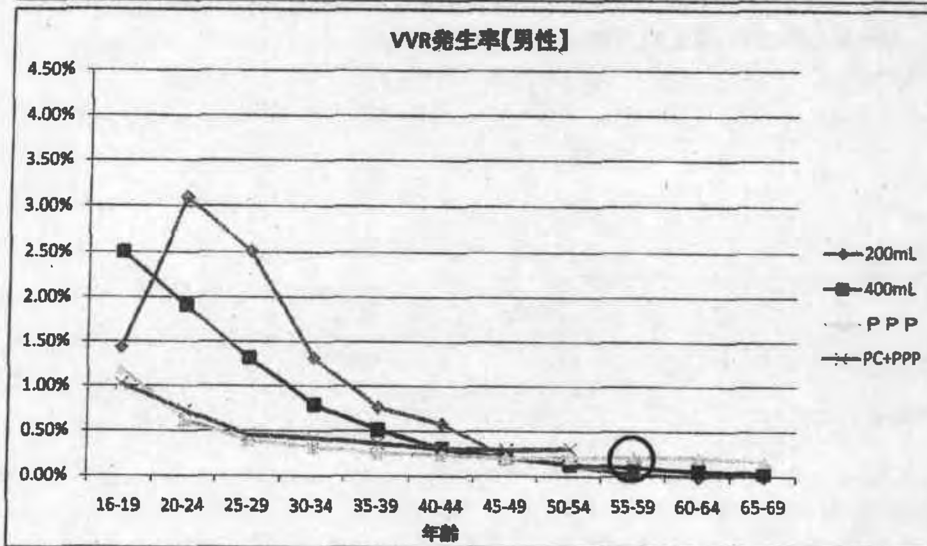


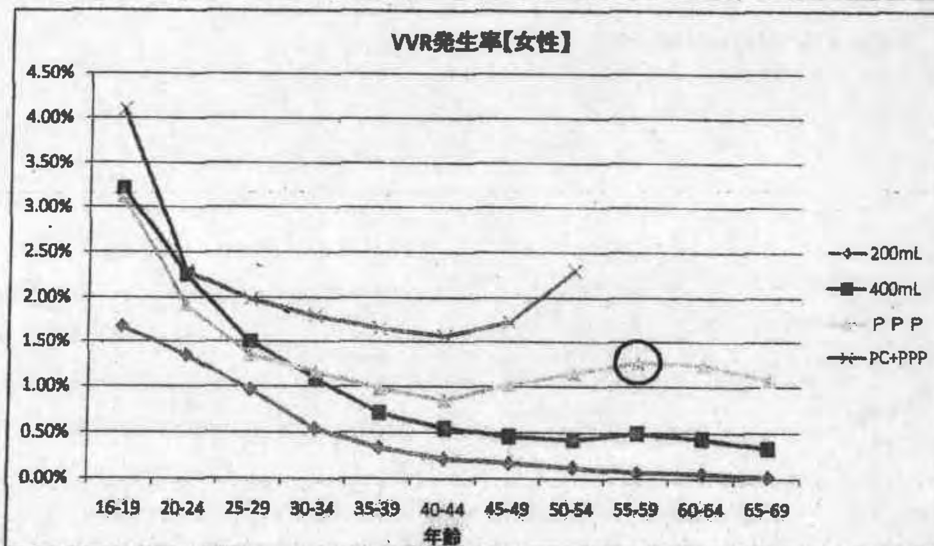




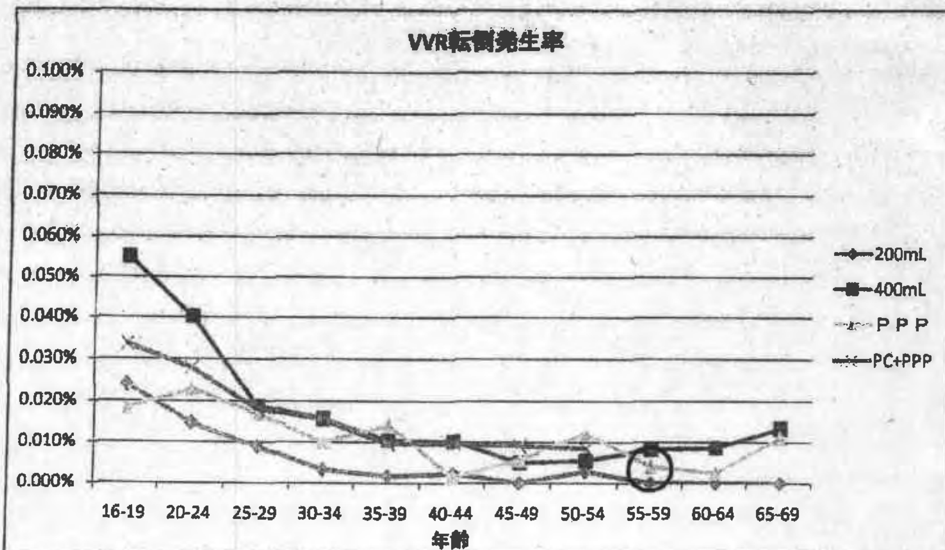
採血副作用発生率(年齢別・性別・採血種類別:平成19年度)

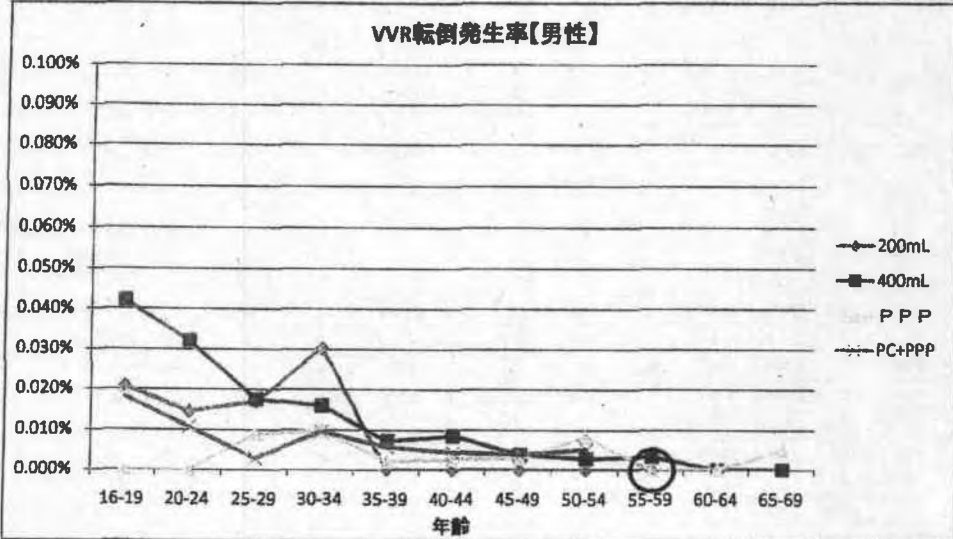


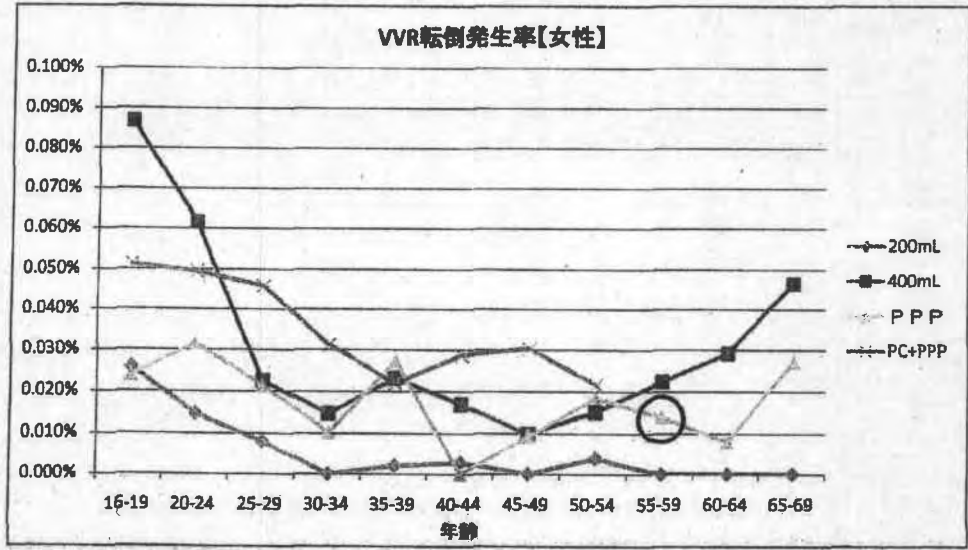




採血副作用発生率(年齢別・性別・採血種類別：平成19年度)





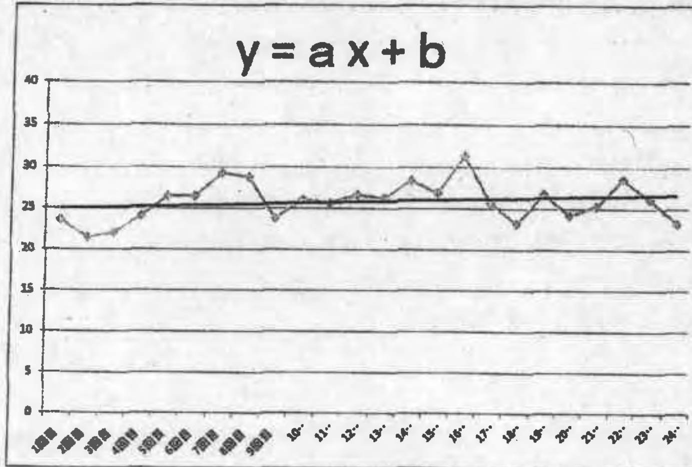


血小板数の推移

対象献血者	：	血小板献血を4年間で24回以上実施した献血者
対象データ	：	追跡開始から24回分のデータ（24回以上でも最初の24回分）

各対象献血者の24回分のデータから回帰直線を作成し、傾き(a)を求めた。

献血回数による血小板数の変化(全献血者)

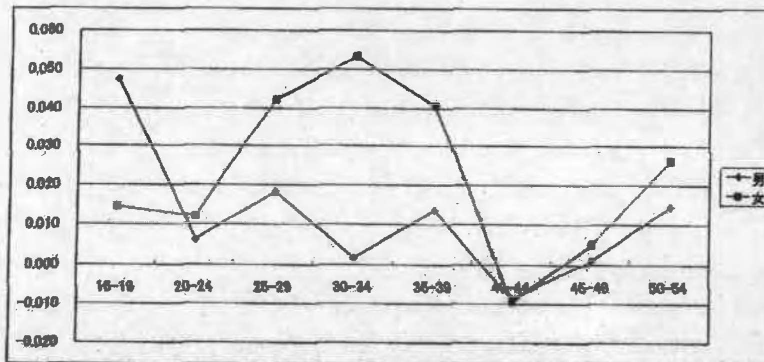


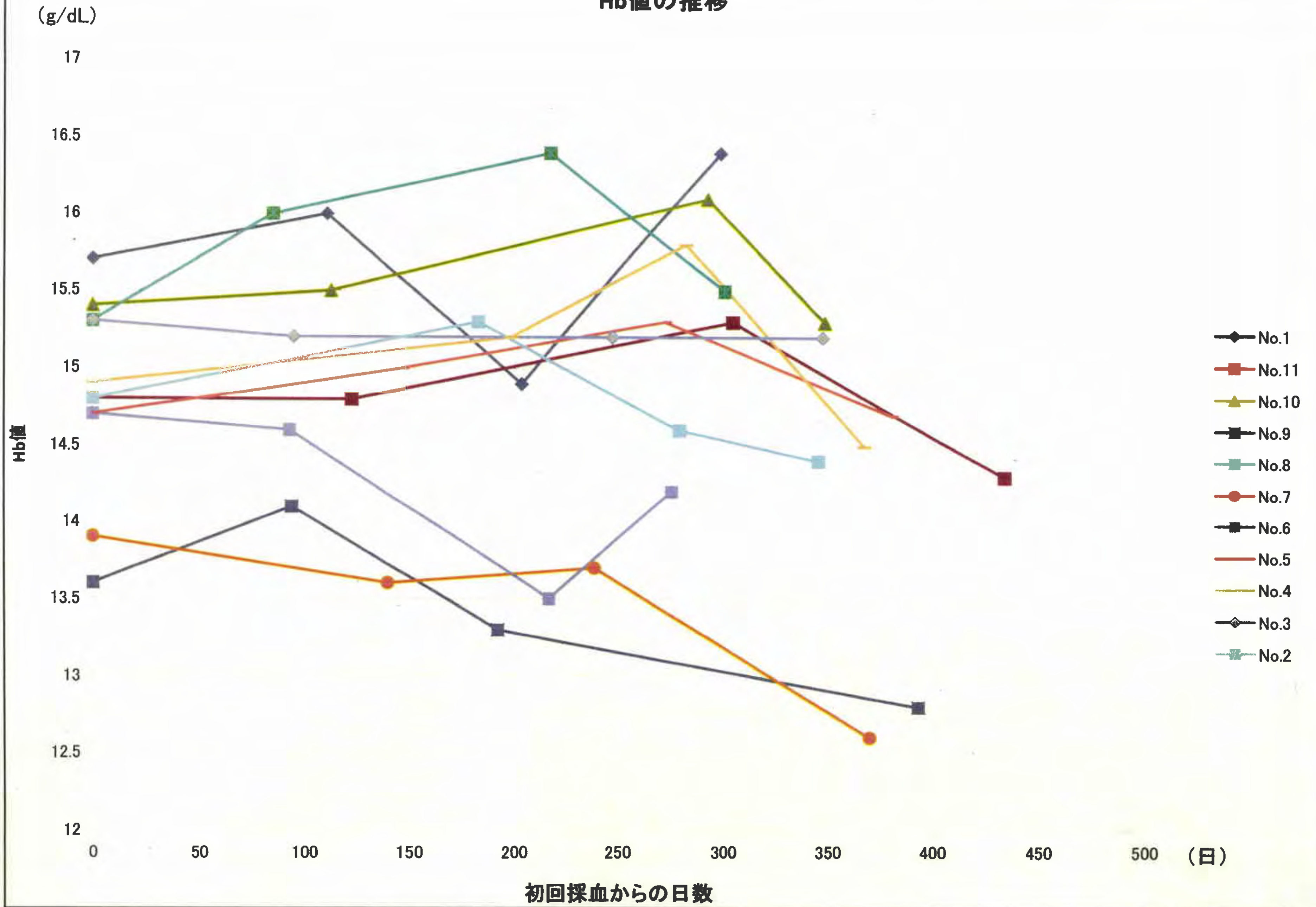
傾きaの加齢による変化

傾き(a)

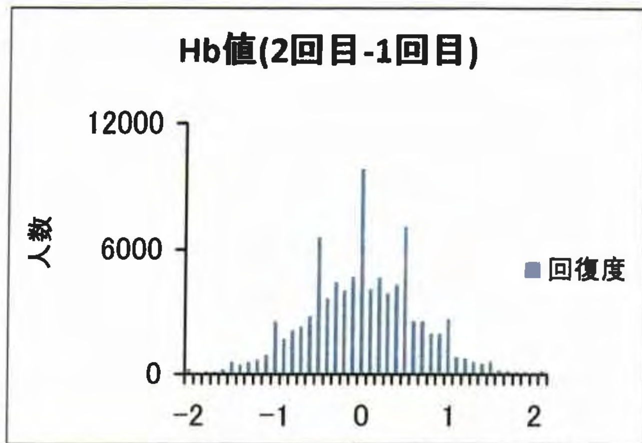
		PT(計)	16-19	20-24	25-29	30-34	35-39	40-44	45-49	50-54
合計	n	2737	88	324	479	538	467	410	321	110
	平均	0.010	0.042	0.007	0.021	0.008	0.016	-0.009	0.001	0.016
	SD	0.102	0.121	0.101	0.099	0.106	0.102	0.096	0.105	0.085
男	n	2403	73	270	414	473	425	364	289	95
	平均	0.007	0.047	0.006	0.018	0.002	0.014	-0.009	0.001	0.014
	SD	0.101	0.117	0.104	0.096	0.102	0.101	0.093	0.106	0.083
女	n	334	15	54	65	65	42	46	32	15
	平均	0.027	0.014	0.012	0.042	0.053	0.041	-0.010	0.005	0.026
	SD	0.111	0.140	0.088	0.110	0.124	0.106	0.116	0.100	0.100

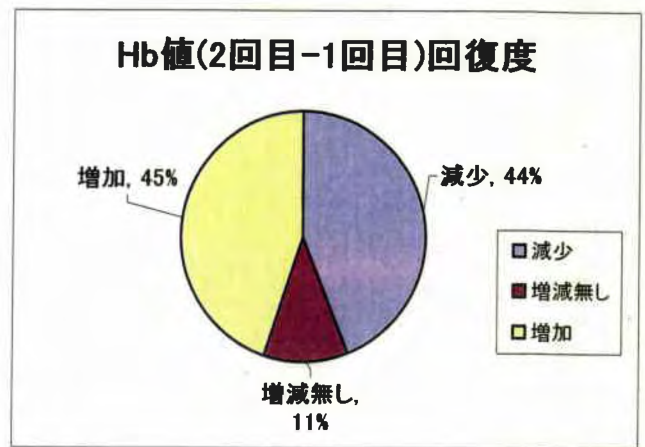
年齢	16-19	20-24	25-29	30-34	35-39	40-44	45-49	50-54
男	0.047	0.006	0.018	0.002	0.014	-0.009	0.001	0.014
女	0.014	0.012	0.042	0.053	0.041	-0.010	0.005	0.026

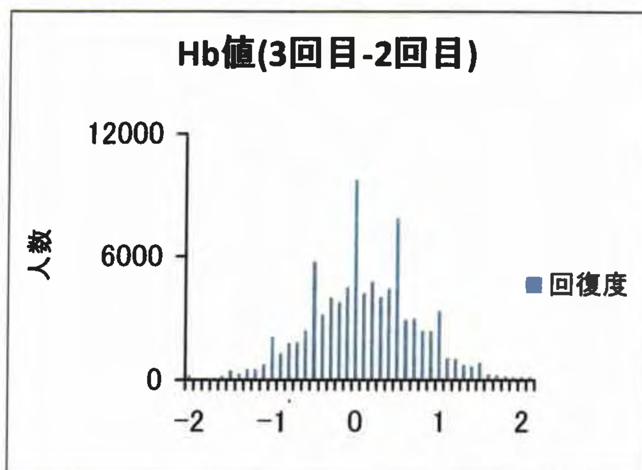


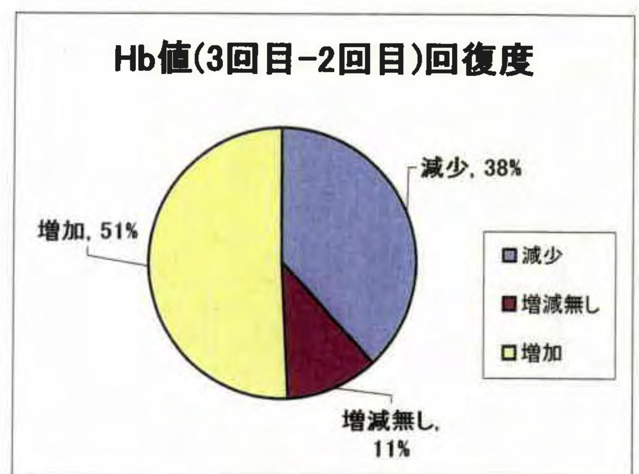


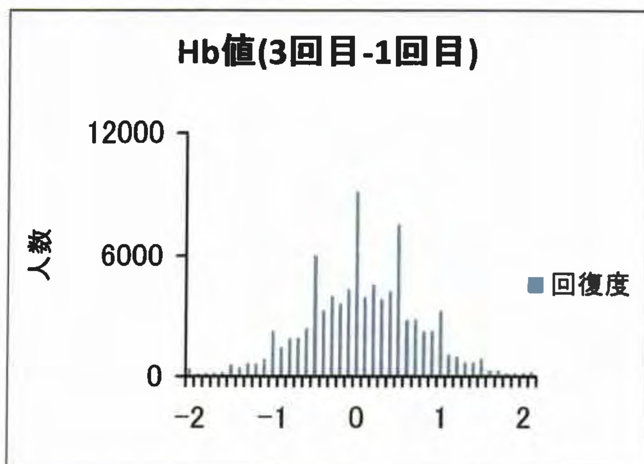
4

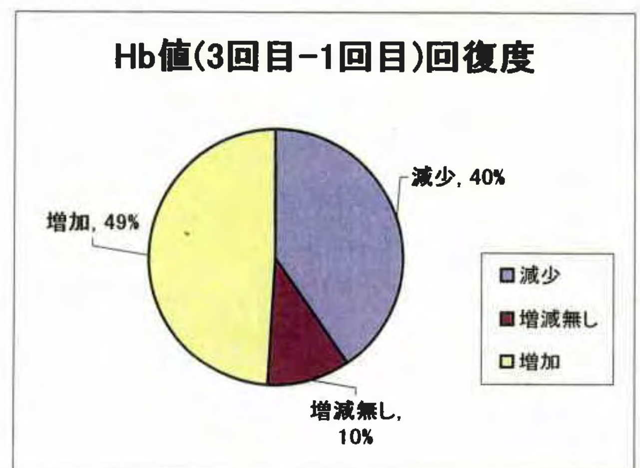

Hb値(2回目-1回目)
12000
6000
0
人数
-2
-1
0
1
2
回復度


Hb値(2回目−1回目)回復度
増加, 45%
減少, 44%
増減無し, 11%
減少
増減無し
増加


Hb値(3回目-2回目)
12000
6000
0
人数
-2
-1
0
1
2
回復度


Hb値(3回目−2回目)回復度
減少, 38%
増加, 51%
増減無し, 11%
減少
増減無し
増加


Hb値(3回目-1回目)
12000
6000
0
人数
-2
-1
0
1
2
回復度


Hb値(3回目−1回目)回復度
減少, 40%
増加, 49%
増減無し, 10%
減少
増減無し
増加

ⅱ　副作用の発生状況については，表4－aの各事項について，採血前，中，後（センター内に留まっている間）について問診し，さらに帰宅時調査用紙（表4－b）を配付して1週間の身体状況について返答を求めた。また，対照として200ml採血者についても同様の調査を行った。

採血前所見の有無と採血中，後，1週間の副作用発生率との関係を男女別にみると，男性群では前所見有りの群でいずれもが，また女性群では前所見有り群の1週間の発生率のみが有意に高率であった。

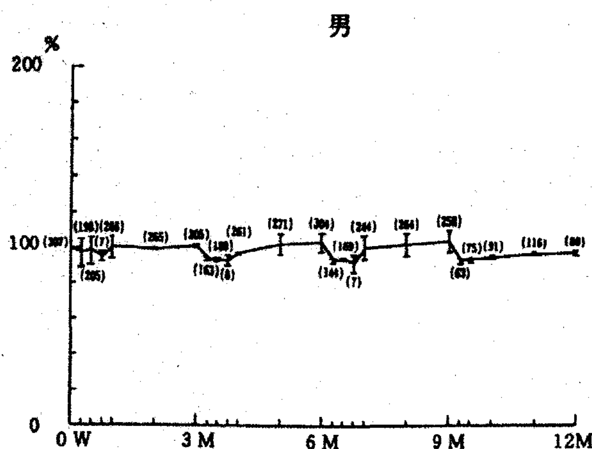
400ml採血後にみられた副作用を，200ml採血（主に移動採血車）後のそれと比較すると，両者間には有意差が認められなかった（表5）。また，母体での200ml採血例に限って検討すると，採血中，1週間の所見には差がみられず，採血後の所見ではむしろ200ml採血例の方が有意に高率であり，また，初回400ml採血例と初回200ml採血例の比較でも採血中，直後の所見は初回200ml採血例の方が有意に高率にみられた。

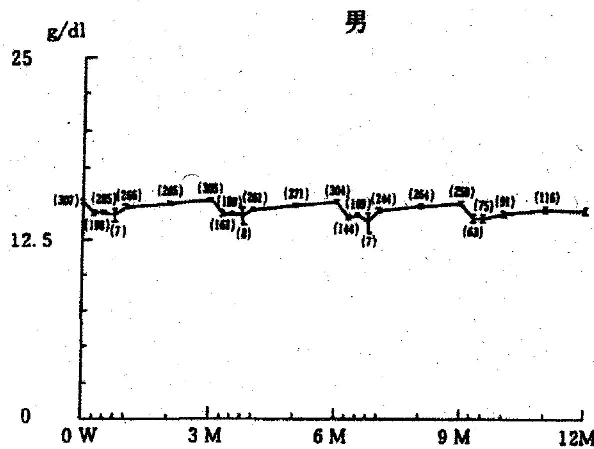
ⅲ　Hb値の回復状況について検討した結果は下記のごとくである。

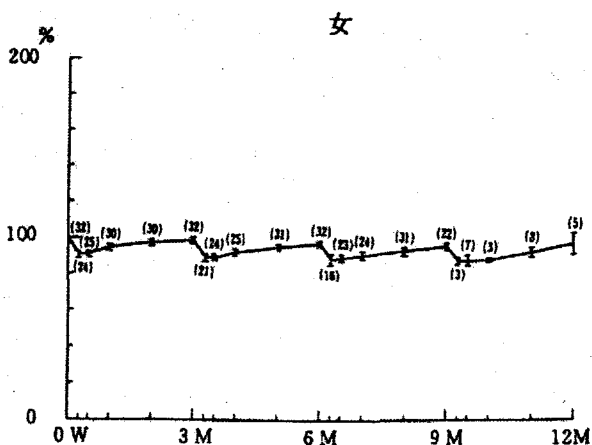
約3か月間隔（3か月±2週間）の採血群中の男性群では，初回採血前値に比して12か月後（4回採血後3か月目）の値は有意（P＜0.001）に低下していた。また，女性群では初回前値と6か月後（2回採血前），初回前値と9か月後（3回採血前）との比較では，それぞれ後者が有意（P＜0.001）に低下していた（図1,2）。約4か月間隔（4か月±2週間）の採血群においては，男女両群ともに採血前値と4か月後(2回採血前）あるいは8か月後（3回採血前）との間には有意差は認められなかった。

ⅳ　血清フェリチン値の回復状況について検討した成績は下記のごとくである。

約3か月間隔の採血群では，男女両群ともに前回







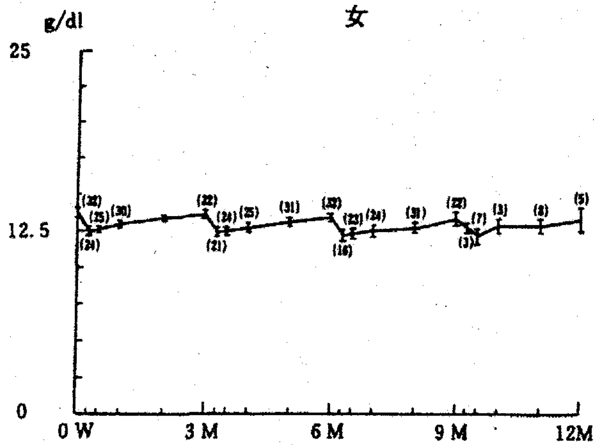


図1　3ケ月間隔採取時のヘモグロビンの回復状況（%）

図2　3ケ月間隔採取時のヘモグロビンの回復状況（g／dℓ）

採血前および採血後1か月毎の検査の平均値（n＝供血者数）

（M ± SD）

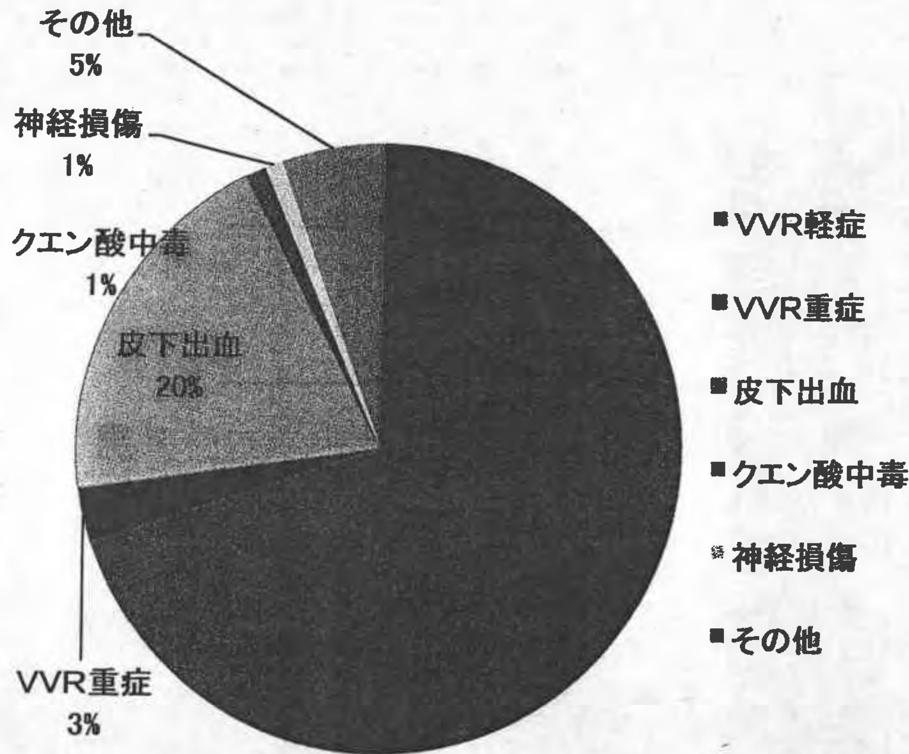
		n	RBC (×10⁴)	Ht (%)	Hb (g/dl)	WBC	Platelet (×10⁴)	Serum Fe (μg/dl)	Ferritin (ng/ml)
0	400 ml 採血前	16	520.6 ± 34.7	46.0 ± 2.7	[illegible]	6562 ± 2072	25.3 ± 5.7	153.5 ± 54.6	70.5 ± 41.2
1	1か月前	15	495.4 ± 30.7	45.3 ± 2.6	15.1 ± 0.7	6020 ± 1544	25.7 ± 4.8	114.8 ± 29.0	42.3 ± 27.6
2	2か月後	15	503.8 ± 27.2	46.1 ± 2.0	15.2 ± 0.5	6020 ± 1299	25.1 ± 4.9	137.0 ± 34.6	43.1 ± 24.3
3	400 ml 採血3か月後	13	495.7 ± 35.4	45.3 ± 2.4	[illegible]	6738 ± 1448	22.5 ± 4.0	155.5 ± 29.4	50.4 ± 23.9
4	4か月後	11	485.4 ± 35.7	44.2 ± 1.5	15.0 ± 0.7	5563 ± 1254	22.0 ± 4.6	144.0 ± 37.8	35.0 ± 19.6
5	5か月後	9	498.7 ± 30.0	45.4 ± 2.4	15.5 ± 0.9	6044 ± 1045	21.7 ± 7.8	129.8 ± 32.4	57.1 ± 31.3
6	400 ml 採血6か月後	11	518.2 ± 25.2	47.0 ± 2.8	[illegible]	5900 ± 1238	25.1 ± 4.2	100.6 ± 18.2	42.3 ± 20.4
7	7か月後	7	496.1 ± 15.4	44.8 ± 2.2	14.9 ± 0.7	6300 ± 1800	23.7 ± 3.4	133.7 ± 51.0	43.5 ± 12.2
8	8か月後	8	518.5 ± 26.0	47.5 ± 2.8	15.5 ± 0.9	6025 ± 796	24.2 ± 7.3	157.3 ± 65.8	36.2 ± 16.0
9	9か月後	7	513.2 ± 17.8	45.8 ± 2.3	15.4 ± 0.7	7626 ± 1411	25.2 ± 4.1	153.4 ± 55.4	35.2 ± 21.0

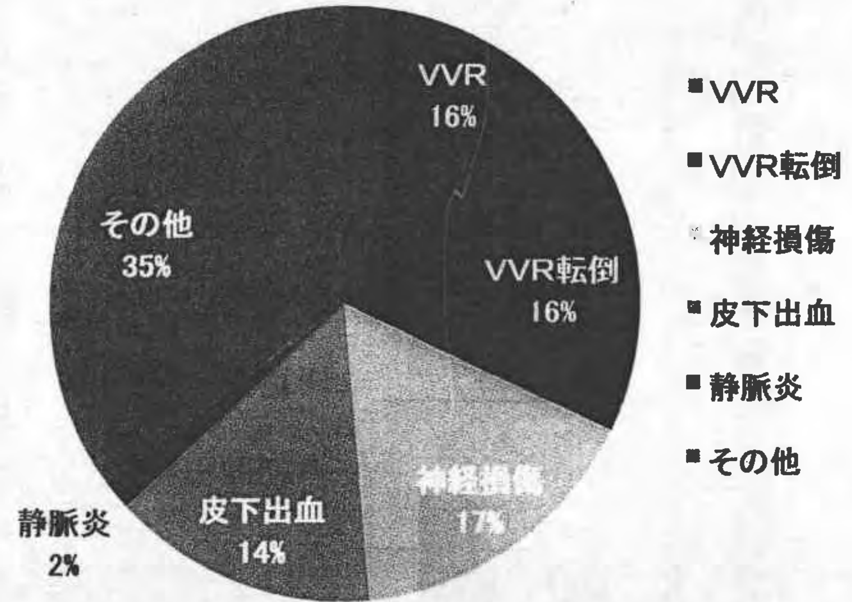
400 ml 採血平均値（男性）

★ P＜0.05

	1回目採血	1 M后	2 M后	2回目採血	1 M后	2 M后	3回目採血	1 M后	2 M后	3 M后	6 M后
血 圧	124±10.2 / 72.5±5.3 n＝4	121.5±20.3 / 78.5±14.8 n＝4	134±14.5 / 94±9.9 n＝4	125±12.9 / 85.5±6.0 n＝4	140±17.8 / 87.3±9.1 n＝4	133±7.4 / 94.5±4.4 n＝4	133±9.3 / 85.5±9.6 n＝4	129±7.6 / 82±2.8 n＝4	123.5±7.9 / 77±11.9 n＝4	130±8.2 / 84±7.1 n＝4	129.5±11.5 / 87±12.1 n＝4
脈 拍		81.8±7.5 n＝4	78±8.5 n＝4	70±10.6 n＝4	85.3±15.5 n＝4	79±6.8 n＝4	76.5±9.0 n＝4	72.5±5.7 n＝4	67.5±9.0 n＝4	73.8±10.0 n＝4	81.0±18.0 n＝4
比 重	1.056 n＝4	1.058±0.0005 n＝4	1.057±0.001 n＝4	1.059+0.0008 n＝4	1.058±0.001 n＝4	1.059 n＝4	1.058±0.001 n＝4	1.059±0.001 n＝4	1.059±0.002 n＝4	1.060 n＝4	1.057±0.0015 n＝4
WBC ×10³	5.45±1.5 n＝4	5.55±0.4 n＝4	5.30±0.4 n＝4	6.0±2.6 n＝4	8.03±1.9 n＝4	5.33±0.8 n＝4	5.88±0.7 n＝4	5.48±1.0 n＝4	5.6±1.2 n＝4	7.13±1.4 n＝4	5.2±0.8 n＝4
RBC ×10⁶	4.945±0.2 n＝4	5.08±0.2 n＝4	4.985±0.3 n＝4	5.12±0.4 n＝4	4.76±0.3 n＝4	5.058±0.3 n＝4	4.715±0.3 n＝4	4.908±0.2 n＝4	4.905±0.3 n＝4	5.18+0.3 n＝4	5.298±0.3 n＝4
Hb g/dl	15.6±0.7 n＝4	15.7±0.8 n＝4	15.2±0.5 n＝4	[illegible] n＝4	14.8±0.8 n＝4	15.7±0.4 n＝4	14.5±0.4 n＝4	15.0±0.5 n＝4	15.2±0.1 n＝4	16.1±0.3 n＝4	16.4±0.5 n＝4
Ht %	44.7±2.0 n＝4	46.4±1.8 n＝4	46.1±0.9 n＝4	47.4±3.3 n＝4	45.1±1.5 n＝4	47.3±1.3 n＝4	44.0±0.6 n＝4	46.1±1.5 n＝4	45.6±0.8 n＝4	★47.5±0.5 n＝4	★49.1±2.3 n＝4
P.LT ×10⁴	25.0±2.5 n＝4	30.4±4.2 n＝4	26.9±1.0 n＝4	26.2±3.4 n＝4	25.4±1.6 n＝4	27.3±3.9 n＝4	28.7±4.0 n＝4	27.4±4.2 n＝4	27.9±3.6 n＝4	26.7±3.2 n＝4	28.1±2.4 n＝4
Fe μg/dl	167±37.9 n＝4	144±41.2 n＝4	167.5±51.0 n＝4	121±15.2 n＝4	177.5±32.6 n＝4	178.5±21.9 n＝4	★112.5±18.1 n＝4	172.8±89.7 n＝4	★107.8±26.7 n＝4	118.3±25.3 n＝4	114.5±21.5 n＝4
鉄結合能 mcg/dl	364±37.2 n＝4	386±30.9 n＝4	396±24.9 n＝4	410±35.4 n＝4	386±38.3 n＝4	416±23.6 n＝4	394±57.5 n＝4	399±51.4 n＝4	396±41.6 n＝4	411±44.7 n＝4	386±39.4 n＝4
フェリチン ng/ml	89.9±32.9 n＝4	59.2±34.7 n＝4	59.2±24.1 n＝4	55.2±22.9 n＝4	★34.6±15.8 n＝4	45.2±26.5 n＝4	★38.7±18.6 n＝4	★19.6±7.2 n＝4	★24.5±13.2 n＝4	★39.3±24.1 n＝4	★41.5±15.0 n＝4

年令 21.3±1.0 才
身長 172±2.0 cm
体重 60.3±1.7 kg





副作用種類	VVR軽症	VVR重症	皮下出血	クエン酸中毒	神経損傷	その他	合計
発生件数	37,257	1,553	10,433	581	469	2,953	53,246
発生率(%)	0.75	0.03	0.01	0.21	0.01	0.06	1.07

＊副作用1～5

副作用種類	VVR	VVR転倒	神経損傷	皮下出血	静脈炎	その他	合計
発生件数	118	114	120	105	12	256	725
発生率(%)	0.002	0.002	0.002	0.002	0.000	0.005	0.015

採血副作用には、本採血前（不採血）の副作用も含む。

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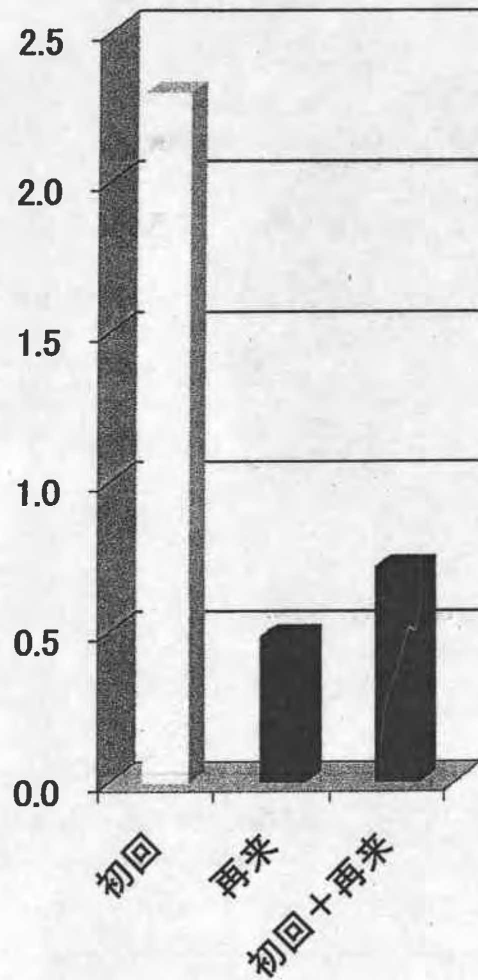
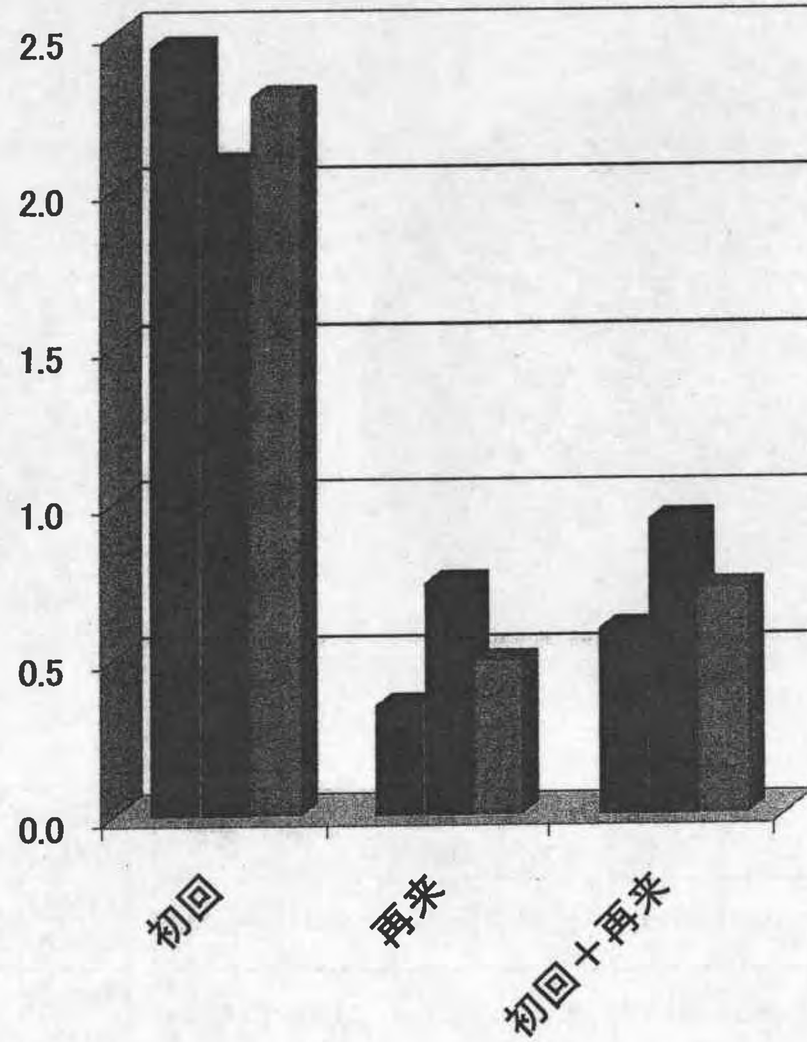
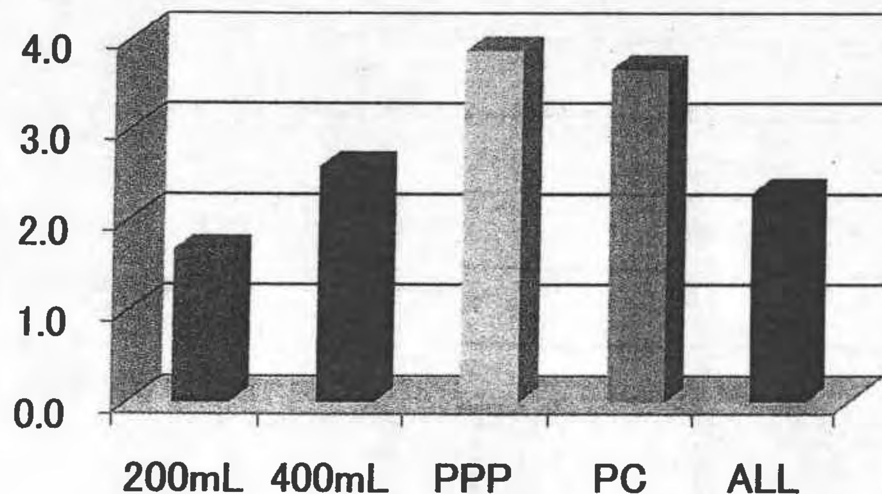
初回·再来とVVR発生率

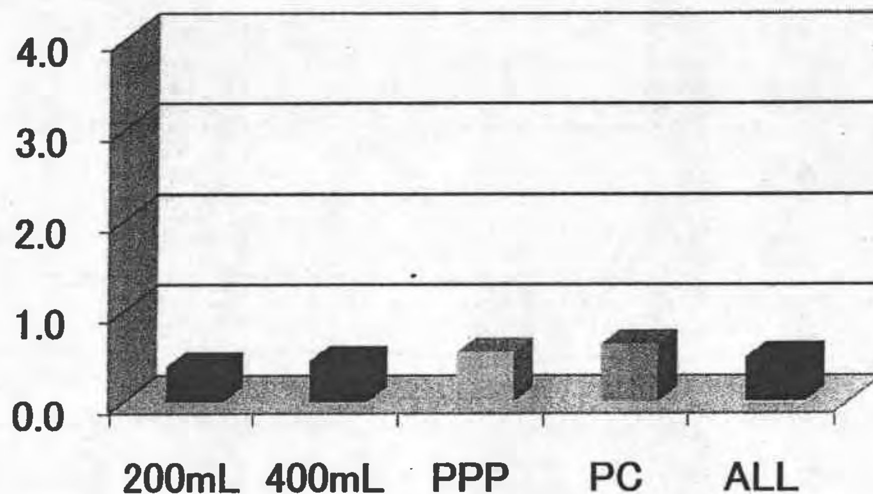


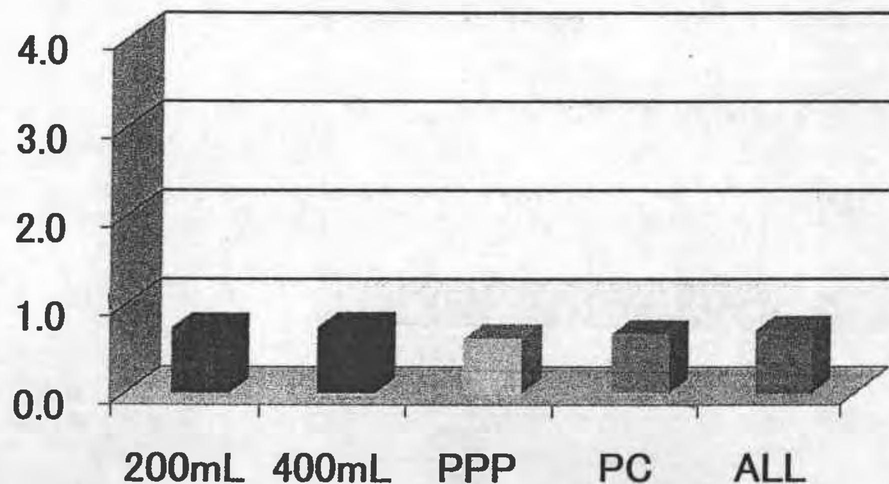


平成18年1月～平成18年12月

Japanese Red Cross Society







200mL
400mL
PPP
PC
ALL

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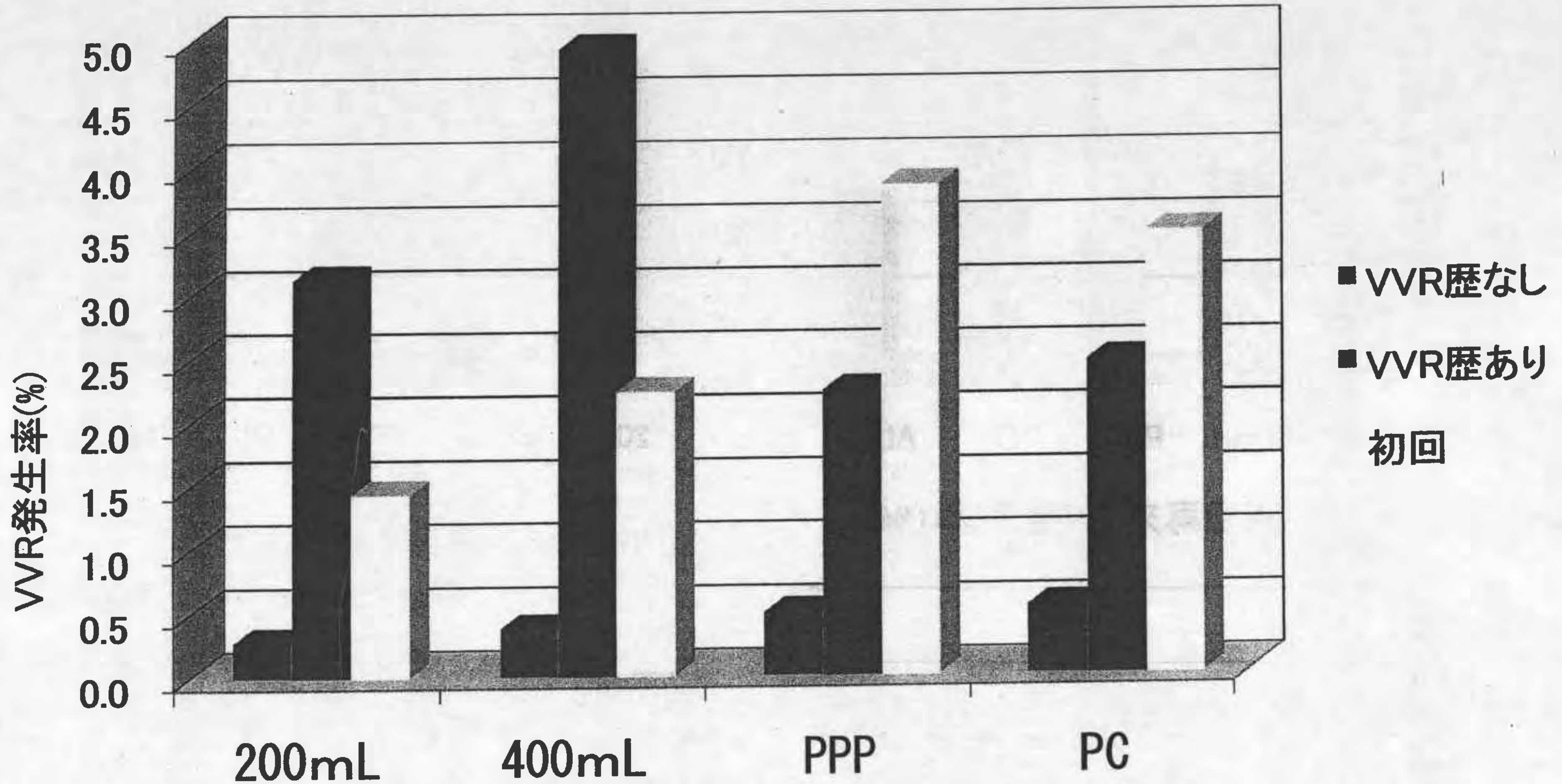
初回献血者、VVR歴あり献血者のVVR発生率(%)



	VVR歴なし	VVR歴あり	初回
200mL	0.28	3.13	1.43
400mL	0.38	4.93	2.23
PPP	0.50	2.24	3.85
PC	0.54	2.46	3.48

平成16年10月～平成17年9月

Japanese Red Cross Society

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Annex 2
Requirements for the collection, processing and quality control of blood, blood components and plasma derivatives (Requirements for Biological Substances No. 27, revised 1992)

Introduction 36

General considerations 37

International Biological Standards and Reference Reagents 39

Definitions 39

Part A. Requirements for the collection of source materials 41

1. Premises 41
2. Equipment 42
3. Personnel 43
4. Donors 43
   - 4.1 Donor selection 43
   - 4.2 Donation frequency and volume 45
   - 4.3 Medical history 46
   - 4.4 Physical examination 48
   - 4.5 Additional requirements applicable to donors for plasmapheresis 49
   - 4.6 Donors for platelet and leukocyte apheresis 51
   - 4.7 Donor immunization and plasma for special purposes 52
5. Collection of blood and plasma 57
   - 5.1 Blood collection and apheresis procedures 57
   - 5.2 Containers 58
   - 5.3 Anticoagulants 59
   - 5.4 Pilot samples 59
   - 5.5 Identification of samples 59

Part B. Requirements for single-donor and small-pool products 60

6. General considerations 60
7. Production and control 60
   - 7.1 General requirements 60
   - 7.2 Testing of whole blood and plasma 61
   - 7.3 Blood-grouping 62
   - 7.4 Red cells 63
   - 7.5 Plasma 66
   - 7.6 Platelets 68
   - 7.7 Leukocytes 70
   - 7.8 Cryoprecipitated factor VIII 71
   - 7.9 Labelling 72

Part C. Requirements for large-pool products 73

8. Introduction 73

9. Buildings 73
- 9.1 Storage of whole blood and plasma 73
- 9.2 Separation of cells and fractionation of plasma 73
- 9.3 Supply and recovery of ancillary materials 74
- 9.4 Viral inactivation 74
- 9.5 Freeze-drying, filling, packaging, labelling and storage 74
- 9.6 Keeping of records 74
- 9.7 Quality control 74
- 9.8 Disposal of infective material 74

10. Equipment 74

11. Provision of support services 75
- 11.1 Water supply 75
- 11.2 Steam supply 75
- 11.3 Other support facilities 75

12. Personnel 76

13. Production control 77
- 13.1 Fractionation of source materials 77
- 13.2 Storage and control of source materials 78

14. Control of albumin and plasma protein fraction 80
- 14.1 Stability of albumin solutions 80
- 14.2 Control of bulk material 81
- 14.3 Control of the final bulk solution 81
- 14.4 Filling and containers 82
- 14.5 Control tests on the final product 83
- 14.6 Records 85
- 14.7 Samples 85
- 14.8 Labelling 85
- 14.9 Distribution and shipping 85
- 14.10 Storage and shelf-life 85

15. Control of immunoglobulins 86
- 15.1 Potency of normal immunoglobulins 87
- 15.2 Potency of specific immunoglobulins 87
- 15.3 Sterility and safety 88
- 15.4 Identity test 88
- 15.5 Freedom from pyrogenicity 89
- 15.6 Moisture content 89
- 15.7 Hydrogen ion concentration 89
- 15.8 Stability 89
- 15.9 Records 90
- 15.10 Samples 90
- 15.11 Labelling 90
- 15.12 Distribution and shipping 90
- 15.13 Storage and shelf-life 90

16. Control of preparations of coagulation-factor concentrates (factor VIII, factor IX and fibrinogen) 91
- 16.1 Tests on final containers 91
- 16.2 Test applicable to factor VIII concentrates 92
- 16.3 Tests applicable to factor IX concentrates 93
- 16.4 Test applicable to fibrinogen 93
- 16.5 Identity test 93
- 16.6 Records 94
- 16.7 Samples 94

16.8 Labelling 94
16.9 Distribution and shipping 94
16.10 Storage and shelf-life 94

Part D. National control requirements 94

17. General 94

18. Release and certification 95

Authors 95

Acknowledgements 96

References 97

Appendix
Summary protocol for collection of source material 99

Introduction

In 1976, a WHO Working Group on the Standardization of Human Blood Products and Related Substances (*1*) considered the need for international requirements for the processing and control of whole human blood and blood products. It emphasized that, as the quality of the source material played an important part in determining the quality of the final products, such requirements should cover all the stages in the process, from the collection of the source materials to the quality control of the final product. In response to the Working Group's recommendations, the Requirements for the Collection, Processing and Quality Control of Human Blood and Blood Products were published in 1978 (*2*). These Requirements were updated and revised in 1988 (*3*), and WHO recommendations concerning testing for antibodies to human immunodeficiency virus (HIV, *4*) were taken into account. This Annex contains a further revision of the Requirements, applicable to the quality control of blood, blood components and plasma derivatives.

A number of other WHO publications have dealt with whole blood and its components, among them guidelines intended mainly for blood transfusion services (*5*). Guidelines of a more general nature, such as the Guidelines for National Authorities on Quality Assurance for Biological Products, have also been published (*6*). The latter call for a quality-assurance system based on the existence of a national structure that is independent of the manufacturer and is responsible for granting licences for biological products, defining procedures for product release and setting up a post-marketing surveillance system. These Guidelines should be followed by any country having or wishing to set up an organization for the collection and fractionation of blood and blood components.

The names of the many experts who provided advice and data taken into account in this revision of the Requirements are listed in the Acknowledgements section, page 96.

General considerations

The setting up of an organization for the collection and fractionation of human blood and blood components calls for a great deal of expertise and considerable investment. Any country contemplating the establishment of such an organization should carry out a careful cost-benefit analysis to determine whether the investment is justified. A logical developmental sequence for a comprehensive organization starts with the collection and distribution of whole blood, progressing later to the separation of whole blood into components and then the fractionation of plasma pools. It is not always possible to be specific about the details of the procedures employed, the in-process controls or the tests applied at each stage of production, in particular for whole blood and component cells. In addition, although the general principle of fractionation of plasma is well established, there are in practice numerous variations in the details of the various production steps. Therefore, any country wishing to begin the collection and fractionation of blood and blood components should send personnel for training to a plant that is operating successfully. WHO may be able to help in arranging such training.

One of the basic questions to be answered by a country considering whether to start fractionation of plasma is whether there is a suitable donor population of sufficient size to guarantee an adequate supply of source material. It is not possible to set a lower limit for the quantity of source material that would be necessary to make such an operation economic because too many factors are involved. However, in order to maintain competence in production and to avoid certain contamination risks, it is important to have sufficient source material to maintain the fractionation facility in continuous operation.

In a comprehensive organization, the greatest expense is that involved in setting up the fractionation plant, but it is also possible to regard the collection of source material and its fractionation as quite separate operations. A country may wish to establish collection centres for separating the cell components and then send the plasma to an established fractionation plant in another country, from where the products could be returned to the original country. The costs of such an operation might be less than those involved in establishing and operating a fractionation plant.

The general prevalence of certain infectious diseases, such as various forms of hepatitis and parasitic diseases, and of HIV infection differs so markedly in different geographical regions that each national authority must decide for itself whether it is cost-effective to apply the most sensitive test to each blood donation and whether it is feasible to collect suitable source material. A brief protocol for the collection of source material is in any case mandatory (see Appendix). Great emphasis should be placed on the production of fractions by a process that experience has shown results in the least risk of contamination. For example, immunoglobulin prepared by the cold ethanol fractionation method of Cohn has a well established

clinical record of being free from contamination with HIV and hepatitis B virus (HBV), as have albumin products prepared by the same method, stabilized and heated for 10 hours at 60 °C (*5*). Nevertheless, extreme care is required in manufacture to ensure that these products are free from infectious viruses, and it cannot be assumed that different fractionation methods will be equally effective. When a fractionation process is introduced or significant modifications are made to an existing production process, the process or the modifications should be validated or revalidated by appropriate procedures, including the use of marker viruses and, where applicable, special *in vitro* and *in vivo* testing.

Blood can harbour a number of different viruses, and the use of medicinal products derived from human blood has led to transmission of viruses such as HBV and HIV. The risk of virus transmission by blood and blood products can be diminished by the testing of all individual donations. Policies for mandatory testing shall be determined by the national control authority, and should be reviewed regularly and modified according to the current state of knowledge.

Special care and appropriate measures approved by the national control authority must be taken to protect the health of the staff of blood collection and fractionation facilities.

The transport of source materials from blood collecting centres and hospitals to fractionation facilities requires special consideration. Refrigeration at the temperature range appropriate for the product must be efficient and reliable and proved to be so by monitoring. Thermal insulation must provide an adequate safeguard against a temporary failure of refrigeration. Containers of liquid source material should be filled so as to minimize frothing due to shaking. Because of the potentially infective nature of these biological materials, suitable protection should be provided against breakage, spillage and leakage of containers.

In these Requirements, the word "human" has been omitted from the names of products derived from human blood. Products of animal origin are immunogenic, and their administration to humans should be avoided whenever equivalent products of human origin can be used instead. The proper name of any blood product of non-human origin should include the species of origin.

These Requirements consist of four parts:

- Part A. Requirements for the collection of source materials
- Part B. Requirements for single-donor and small-pool products
- Part C. Requirements for large-pool products
- Part D. National control requirements.

Each deals with a separate aspect of collection, processing and quality control, but all the parts are intended to be taken together to constitute a single document. It will not be possible to rely on any blood product unless the relevant requirements for each step are complied with, and any attempt

to make them less stringent may have serious consequences for the safety of the final product.

Parts A-D are divided into sections, each of which constitutes a recommendation. The parts of each section printed in normal type have been written in the form of requirements, so that, if a health administration so desires, they may be adopted as they stand as definitive national requirements. The parts of each section printed in small type are comments or recommendations for guidance.

Should individual countries wish to adopt these Requirements as the basis for their national regulations concerning blood products and related substances, it is recommended that modifications be made only on condition that the modified requirements ensure at least an equal degree of safety and potency of the products. It is desirable that the World Health Organization should be informed of any such changes.

Increasing demand for blood products is resulting in the extensive movement of such products from one country to another. Internationally accepted requirements are therefore necessary so that countries without any regulations on blood products and related substances may refer to them when importing such products.

International Biological Standards and Reference Reagents

Rapid technological developments in the measurement of the biological activity of blood products and related substances require the establishment of international biological reference materials. The first two such materials (for anti-A and anti-B blood-typing sera) were established in 1950, and further reference materials have been established since. A number of materials are currently under investigation for use in the preparation of new standards.

The activity of blood products must be expressed in International Units where an International Standard exists. WHO publishes a list of such standards (revised from time to time and most recently in 1990) under the title *Biological substances: International Standards and Reference Reagents.*

Definitions

The following definitions are intended for use in this document and are not necessarily valid for other purposes.

Blood collection: a procedure whereby a single donation of blood is collected in an anticoagulant and/or stabilizing solution.

Processing: any procedure that takes place after the blood is collected.

Plasmapheresis, apheresis and cytapheresis: procedures whereby whole blood is separated by physical means into components and one or more of them returned to the donor.

Closed blood-collection and processing system: a system for collecting and processing blood in containers that have been connected together by the manufacturer before sterilization, so that there is no possibility of bacterial or viral contamination from outside after collection of blood from the donor.

Donor: a person who gives blood or one of its components.

Single-donor materials

Whole blood (sometimes referred to as "blood"): blood collected in an anticoagulant solution with or without the addition of nutrients such as glucose or adenine. Whole blood is collected in units of 450 ml.

Blood component: any part of blood separated from the rest by means of physical procedures.

Plasma: the liquid portion remaining after separation of the cellular elements from blood collected in a receptacle containing an anticoagulant, or separated by continuous filtration or centrifugation of anticoagulated blood in an apheresis procedure.

Plasma, frozen: a plasma separated more than 8 h after collection of the blood and stored below -20 °C.

Plasma, fresh-frozen: a plasma separated within 8 h of donation, frozen rapidly and stored below -20 °C (and preferably below -30 °C).

Plasma, platelet-rich: a plasma containing at least 70% of the platelets of the original whole blood.

Plasma, freeze-dried: any one of the above forms of plasma that has been freeze-dried for preservation.

Plasma, recovered: plasma recovered from a whole blood donation.

Cryoprecipitated factor VIII: a crude preparation containing factor VIII that is obtained from single units (or small pools) of plasma derived either from whole blood or by plasmapheresis, by means of a process involving freezing, thawing and precipitation.

Serum: the liquid part of coagulated blood or plasma.

Red cells: whole blood from which most of the plasma has been removed and having an erythrocyte volume fraction greater than 0.7.

Red cells suspended in additive solution: red cells to which a preservative solution, for example containing adenine, glucose and mannitol, is added to permit storage for longer periods; the resulting suspension has an erythrocyte volume fraction of approximately 0.6-0.7.

Red cells, washed: red cells from which most of the plasma has been removed by one or more stages of washing with an isotonic solution.

Red cells, leukocyte-depleted: a unit of a red-cell preparation containing fewer than 1.2×10^9 leukocytes.

Red cells, leukocyte-poor: a unit of a red-cell preparation containing fewer than 5×10^6 leukocytes.

Red cells, frozen: red cells that have been stored continuously at −65 °C or below, and to which a cryoprotective agent such as glycerol has been added before freezing.

Red cells, deglycerolized: frozen red cells that have been thawed and from which glycerol has been removed by washing.

Platelets: platelets obtained either by separation of whole blood, buffy coat or platelet-rich plasma or by apheresis and suspended in a small volume of plasma from the same donation.

Leukocytes: leukocytes obtained either by the separation of whole blood or by apheresis and suspended in a small volume of plasma from the same donation.

Large-pool products

Bulk material: plasma, powder, paste or liquid material prepared by the fractionation of pooled plasma.

Final bulk: a sterile solution prepared from bulk material and bearing the corresponding batch number. It is used to fill the final containers.

In some countries, the final bulk is distributed into containers through a sterilizing filter. If the total final bulk is not distributed into containers in one session, each of the filling lots is given a sub-batch number.

Filling lot (final lot): a collection of sealed final containers that are homogeneous with respect to composition and the risk of contamination during filling and (where appropriate) drying or other further processing such as heat treatment. A filling lot must therefore have been filled and (where appropriate) dried in one working session.

Part A. Requirements for the collection of source materials

1. Premises

The premises shall be of suitable size, construction and location to facilitate their proper operation, cleaning and maintenance in accordance with accepted rules of hygiene. They shall comply with the requirements of Good Manufacturing Practices for Pharmaceutical (*7*) and Biological (*8*) Products and in addition provide adequate space, lighting and ventilation for the following activities where applicable:

- The medical examination of individuals in private to determine their fitness as donors of blood and/or blood components and to provide an opportunity for the confidential self-exclusion of unsuitable potential donors.
- The withdrawal of blood from donors and, where applicable, the re-infusion of blood components with minimum risk of contamination and errors.
- The care of donors, including the treatment of those who suffer adverse reactions.
- The storage of whole blood and blood components in quarantine pending completion of processing and testing.
- The laboratory testing of blood and blood components.
- The processing and distribution of whole blood and blood components in a manner that prevents contamination and loss of potency.
- The performance of all steps in apheresis procedures, if applicable.
- The performance of labelling, packaging and other finishing operations in a manner that prevents errors.
- The storage of equipment.
- The separate storage of quarantined and finished products.
- The documentation, recording and storage of data on the donor, the donated blood and the ultimate recipient.

Mobile teams can be used for the collection of blood. Although the premises used by such teams may not comply with the more stringent requirements for centres built specially for the purpose, they must be adequate to ensure the safety of the donor, the collected blood or blood components and the staff participating in blood collection. The safety of the subsequent users of the premises should also not be forgotten.

2. Equipment

The equipment used in the collection, processing, storage and distribution of blood and blood components shall be calibrated, tested and validated before initial use, and shall be kept clean and maintained and checked regularly. The requirements of Good Manufacturing Practices for Pharmaceutical (*7*) and Biological (*8*) Products shall apply in every particular.

The equipment employed to sterilize materials used in the collection of blood or blood components or for the disposal of contaminated products shall ensure that contaminating microorganisms are destroyed and shall be validated for this purpose. The effectiveness of the sterilization procedure shall be not less than that achieved by a temperature of 121.5 °C maintained for 20 min by means of saturated steam at a pressure of 103 kPa (1.05 kgf/cm^2 or 15 lbf/in^2) or by a temperature of 170 °C maintained for 2 h with dry heat.

All contaminated material should be made safe before disposal. Disposal should comply with the relevant local laws.

Tests for sterility are given in the revised Requirements for Biological Substances No. 6 (General Requirements for the Sterility of Biological Substances) (9, pp. 40–61).

3. Personnel

An organization for the collection of blood or blood components shall be under the direction of a designated and appropriately qualified person who shall be responsible for ensuring that all operations are carried out properly and competently. The director shall have adequate knowledge and experience of the scientific and medical principles involved in the procurement of blood and, if applicable, the separation of blood components and the collection of such components by apheresis.

The director shall be responsible for ensuring that employees are adequately trained and acquire practical experience and that they are aware of the application of accepted good practice to their respective functions.

The director should have the authority to enforce or to delegate the enforcement of discipline among relevant employees.

The persons responsible for the collection of the blood and blood components shall be supervised by licensed physicians who shall be responsible for all medical decisions, for review of the procedures manual and for the quality-control programme, including techniques, equipment, procedures and staff.

The personnel responsible for the processing, storage, distribution and quality control of blood, blood components and plasma shall be adequate in number and each member of the personnel shall have a suitable educational background and training or experience that will ensure competent performance of assigned functions so that the final product has the required safety, purity, potency and efficacy.

4. Donors

4.1 *Donor selection*

The provision of blood, blood components and plasma derivatives from voluntary, non-remunerated donors should be the aim of all countries.

In selecting individuals for blood donation, it is most important to determine whether the person is in good health, in order to protect the donor against damage to his or her own health and to protect the recipient against exposure to diseases or to medicinal products from the blood or blood products. It should be recognized that the donor selection process contributes significantly to the safety of blood products derived from large plasma pools. The following provisions apply to donations of blood or blood components not intended for autologous use.

The health of a donor shall be determined by a licensed physician or a person under the direct supervision of a licensed physician, and the donor shall be free from any disease transmissible by blood transfusion in so far as can be determined by history-taking and examination (see section 4.3). Donors shall be healthy persons of either sex between the ages of 18 and 65 years.

In some countries, there is no upper limit to the age of the donor. With parental consent the minimum age may be lowered to 16 years.

Red blood cells from donors with glucose-6-phosphate dehydrogenase deficiency, sickle-cell trait or other inherited erythrocyte abnormalities may give rise to transfusion reactions under certain circumstances. Decisions regarding the suitability of such donors should be made by the national control authority.

A donor should be considered for plasmapheresis only where the procedures involved result in products or services shown to serve accepted medical purposes, including prophylaxis, therapy and diagnosis, as verified by valid scientific evidence. All donors should be certified as acceptable, at the time of each plasmapheresis procedure, by a registered physician or by trained personnel under the direct supervision of the physician.

Those eligible for apheresis donation include: (a) healthy persons who fulfil the general criteria for blood donors; (b) persons with antibody levels that have been increased, either naturally or by immunization; (c) subject to (a) above, persons with plasma that is of value for diagnostic or reference purposes; and (d) persons whose blood may be used in the preparation of certain vaccines.

When a potential donor does not fulfil the general criteria for blood donation, the acceptance of her or him as a donor for a specific component of blood should be at the discretion of the responsible physician. Where appropriate, the physician should have access to an ethical committee.

Donor education and selection programmes are intended to prevent potentially infectious units of blood and plasma from being collected. It is essential that such programmes are comprehensible and readily accessible to all potential donors.

To reduce the likelihood of transmitting infections, all potential donors should be informed of factors in their history or behaviour that may increase their risk of being infected. The national control authority must determine the appropriate exclusion criteria for the country concerned.

Persons in the following categories shall be excluded from acting as donors:

- those with clinical or laboratory evidence of infectious disease, e.g. infection with hepatitis viruses, HIV-1 or HIV-2;
- past or present intravenous drug abusers;
- men who have had a sexual relationship with another man;

- men and women who have engaged in prostitution;
- those with haemophilia or other clotting-factor defects who have received clotting-factor preparations;
- sexual partners of any of the above.

In some countries, the sexual partners of those at risk of transmitting infections are excluded from acting as donors for only one year.

Persons who have received blood transfusions should be excluded from acting as donors for at least one year.

Donors should be made aware before donating blood that it will be tested for the presence of serological markers of infection. It is advisable that the right to test donations and the legal implications of testing donations should be clarified by the appropriate authority.

4.2 *Donation frequency and volume*

4.2.1 *Whole blood*

The frequency of whole-blood donations shall not exceed once every two months, with a maximum volume in any consecutive 12-month period of 3 l.

A standard donation should not be collected from persons weighing less than 50 kg.

A *standard donation* is 450 ml; an optimum blood/anticoagulant ratio is 7 to 1.

The frequency of donation may have to be modified on an individual basis. In general, premenopausal women should not donate blood as frequently as men.

4.2.2 *Plasma*

Plasma donors can be divided into three groups: those who donate at a frequency comparable to that allowed for whole-blood donations; those who donate two to three times as frequently as whole-blood donors; and those who donate at a maximum of twice a week. The first group shall be accepted on the basis of the general criteria for blood donors.

The maximum volume of plasma that may be removed from a donor during one plasmapheresis procedure shall be determined by the national health authority, and shall depend on whether the plasma is obtained by manual or automated plasmapheresis.

In some countries, the volume of plasma collected during a manual procedure is the quantity obtained from 1.0–1.2 l of whole blood. The volume of plasma collected during an automated procedure depends on the equipment used.

It is difficult to specify the maximum volumes of plasma that can be safely collected from donors until more definitive data are available on the effects of plasmapheresis on donors. The limits imposed in different countries vary, and depend on the nutritional status of the donor.

If a plasma donor donates a unit of whole blood or if the red blood cells are

not returned in an apheresis procedure, the next donation shall be deferred by eight weeks unless special circumstances warrant approval by the responsible physician of plasmapheresis at an earlier date.

In general, plasma collected by therapeutic plasmapheresis shall not be used for fractionation.

4.3 *Medical history*

4.3.1 *General*

Before each donation, questions shall be asked so as to ensure that the donor is in normal health and has not suffered, or is not suffering, from any serious illness.

A donor who appears to be suffering from symptoms of acute or chronic disease or who is receiving oral or parenteral medication, with the exception of vitamins, postmenopausal hormone therapy or oral contraceptives, shall not be accepted unless approved by a physician.

A donor who appears to be under the influence of any drug including alcohol or who does not appear to be providing reliable answers to medical history questions shall not be accepted.

4.3.2 *Infectious diseases*

Potential donors with a history that places them at increased risk of transmitting infection shall not donate blood or plasma for an appropriate time period. A donor shall be permanently excluded if one of his or her previous blood donations was believed to be responsible for transmitting disease.

In most countries, questions concerning the signs and symptoms of HIV infection will be part of the routine assessment of medical history and appropriate monitoring for HIV, as defined by the national control authority, will be included. As a result of this assessment, a potential donor may be disqualified.

Donors shall not have a history of: positive laboratory test results for hepatitis or corresponding symptoms and signs; close contact with an individual with hepatitis within the previous year; receipt within the previous year of human blood or any blood component or fraction that might be a source of transmission of infectious agents; or tattooing, scarification or ear piercing (unless performed under sterile conditions) within the previous year.

Acupuncture within the previous year may also present a risk if not carried out under sterile conditions.

In some countries, potential donors with a history of viral hepatitis or of a positive test for hepatitis B surface antigen (HBsAg) or antibodies to hepatitis C virus (anti-HCV) are permanently excluded. In others, such donors are accepted providing that recovery occurred more than one year previously and that the reaction for HBsAg and anti-HCV in a sensitive test is negative.

The requirements concerning viral hepatitis may be varied, at the discretion of the national control authority, according to the local epidemiological circumstances.

The collection both of single-donor products (whole blood and its components) and of plasma for pooling for the manufacture of plasma fractions capable of transmitting hepatitis or HIV should be avoided if a group of potential donors shows a prevalence of acute or chronic hepatitis B, hepatitis C or HIV infection higher than that found in the general donor population. Specific approval may be given by national control authorities for the use of donations from such populations to provide plasma for the manufacture of hepatitis B vaccine or hepatitis B immunoglobulin.

In areas with a low incidence of transfusion-transmitted disease, whole blood or blood components should not be used for transfusion if obtained from source material collected in an area where there is a high incidence of blood-borne infectious disease.

Blood and plasma shall be tested for the presence of HBsAg, anti-HIV and anti-HCV by the methods described in Part B, section 7.2; the tests used should be approved by the national control authority or other appropriate authority.

Anyone whose blood has been shown to be reactive for infectious disease markers by approved screening tests shall be excluded as a donor. Selection as a donor may later be permitted if sufficient data are available from tests approved by the national control authority to indicate that the original results were non-specific.

National health authorities shall develop policies designed to prevent the transmission of infectious diseases based on the prevalence of these diseases in the donor population and the susceptibility of recipients to them.

In countries where malaria is not endemic, donors of cellular blood products should have a negative history of malaria exposure during the previous six months and a negative history of clinical malaria, or a history of malaria prophylaxis if they have resided in, or visited, an endemic area within the three years preceding the donation. Such restrictions may be less important in countries where the prevalence of endemic malaria is high among both donors and recipients, except when blood products are required by visitors from non-endemic areas. Malaria history is not pertinent to plasma donation for source material that will be fractionated.

Particular attention should be paid to skin decontamination procedures before blood collection.

Many parasitic, bacterial and viral diseases, including *trypanosomiasis*, toxoplasmosis, syphilis and brucellosis, can be transmitted by blood. Precautions shoud be taken to avoid blood collection during the viraemic phase of viral diseases like measles and rubella. Potential donors who have lived in or recently travelled to areas where human T-cell lymphotropic virus infections and haemorrhagic fever are endemic should be investigated for evidence of such infections.

Anyone who has received pituitary hormones of human origin should be permanently excluded as a donor because of possible infection with the agent causing Creutzfeldt-Jakob disease, although transmission of this agent through blood products has not been proved.

4.3.3 *Minor surgery*

Donors shall not have undergone tooth extraction or other minor surgery during a period of 72 h before donation.

4.3.4 *Pregnancy and lactation*

Pregnant women shall be excluded from blood donation. In general, mothers shall also be excluded during lactation and for at least six months after full-term delivery.

The interval before blood donation is permissible after pregnancy may be shorter in some cases, e.g. six weeks after an abortion during the first trimester.

In some countries, donors are accepted when pregnant or during the period of lactation if their blood contains certain blood-group antibodies or is needed for autologous transfusion. The volume to be taken should be determined by the physician responsible.

4.3.5 *Prophylactic immunization*

Symptom-free donors who have recently been immunized may be accepted with the following exceptions:

- Those receiving attenuated vaccines for measles, mumps, yellow fever or poliomyelitis shall be excluded until two weeks after the last immunization or injection.
- Those receiving attenuated rubella (German measles) vaccine shall be excluded until four weeks after the last injection.
- Those receiving rabies vaccine for post-exposure treatment shall be excluded until one year after the last injection.
- Those receiving passive immunization with animal serum products shall be excluded until four weeks after the last injection.
- Those receiving hepatitis B vaccine need not be excluded unless the vaccine is being given because of exposure to a specific risk, in which case the donor shall be disqualified for at least 12 months after the last such exposure. If hepatitis B immunoglobulin has been administered, the period of deferral shall be at least 12 months because disease onset may be delayed.

4.4 **Physical examination**

As determined by the national control authority, physical examination of donors may include measurement of weight, blood pressure, pulse rate and temperature. If these are measured and the results lie outside the ranges recommended below, the donor concerned shall be accepted only if approved by the licensed physician in charge.

- *Blood pressure:* systolic blood pressure between 12 and 24 kPa (90 and 180 mmHg); diastolic blood pressure between 6.67 and 13.3 kPa (50 and 100 mmHg).
- *Pulse:* between 50 and 110 beats per minute and regular. Lower values may be accepted in healthy athletes with endurance training.
- *Temperature:* oral temperature not exceeding 37.5 °C.
- *Weight:* donors weighing less than 50 kg may donate a volume of blood proportionally less than 450 ml in an appropriate volume of anticoagulant, provided that all other donor requirements are met.

Donors shall be free from any infectious skin disease at the venepuncture site and of skin punctures or scars indicative of abuse of intravenous drugs.

4.5 Additional requirements applicable to donors for plasmapheresis

All phases of apheresis, including explaining to donors what is involved in the process and obtaining their informed consent, should be performed under the direct supervision of a licensed physician or by trained personnel reporting to such a physician.

4.5.1 *First-time plasma donors*

When prospective plasma donors present themselves to a centre for the first time, initial screening shall begin only after the procedure of plasmapheresis has been explained and the donor has given consent.

The following information shall be permanently recorded:

- Personal information and identification. If the donor is to participate in an ongoing programme, an effective means of identification is especially important. The use of identity numbers, photographs or other equally effective measures should be considered.
- A preliminary medical history as required for blood donors, covering infectious diseases and the donor's general state of health.

If there are no contraindications to plasmapheresis, preliminary laboratory tests shall be carried out, namely reading of the erythrocyte volume fraction or haemoglobin concentration, determination of total serum protein and screening for protein and sugar in the urine. The haemoglobin concentration or erythrocyte volume fraction of the donor's blood shall be within normal limits, as defined by the national control authority or the national blood transfusion authority.

Many countries specify minimum haemoglobin concentrations of 125 g/l for women and 135 g/l for men, or, for microhaematocrit determinations, minimum erythrocyte volume fractions of 0.38 for women and 0.41 for men.

If normal values are also obtained in the other laboratory tests, evaluation of the potential donor by the physician begins.

In some countries, specially trained non-physicians are permitted to conduct these routine examinations under the supervision of a physician.

Donors participating in a programme in which plasmapheresis is more frequent than is blood donation for those eligible for whole-blood collection shall be examined by a licensed physician on the day of the first donation, or not more than one week before that donation. This examination shall include measurement of temperature and blood pressure, auscultation of the heart and lungs, palpation of the abdomen, assessment of neurological signs, urine analysis and blood sampling for tests required by the national control authority. Liver function tests (e.g. for alanine aminotransferase), tests for HBsAg, anti-HIV and anti-HCV, and quantification of plasma proteins by electrophoresis or another suitable method shall also be included. The physician shall obtain informed consent after explaining the procedure of plasmapheresis and describing the hazards and adverse reactions that may occur. At this stage, donors shall be given an opportunity to refuse participation. If they consent, it must be on the condition that their legal rights to recover damages are not waived.

In some countries, the first plasmapheresis procedure may be performed before the results are available for the liver function tests, the serological tests for syphilis (if required by the national control authority) and the tests for HBsAg, anti-HCV and anti-HIV. The results of the tests for quantifying plasma proteins should be reviewed by the physician before subsequent plasmapheresis procedures.

4.5.2 *Donors who have undergone plasmapheresis previously in the same programme*

For donors who have already taken part in a plasmapheresis programme:

- The receptionist shall note the date of the last donation (at least two days must have elapsed since that time). No more than two donations shall be permitted within a seven-day period.
- The medical history and weight of the donor shall be recorded; blood pressure, temperature, pulse rate and haemoglobin concentration shall be measured by trained personnel. On the day of each donation, in addition to meeting the general requirements for donors, plasma donors shall be shown to have a total serum protein concentration of not less than 60 g/l.

The medical evaluation of plasma donors shall be repeated at regular intervals, as specified by the national control authority, and tests carried out as specified in section 4.5.3.

Whenever the result of a laboratory test is found to be outside the established normal limits or a donor exhibits any important abnormalities of history or on physical examination, the donor shall be excluded from the programme. The donor shall not be readmitted to the programme until the results of relevant tests have returned to normal and the responsible physician has given approval in writing. It is the responsibility of national health authorities to define normal ranges and standard deviations of test results on the basis of data from a sufficiently large sample of healthy individuals not undergoing plasmapheresis.

In the case of hepatitis C, the results of liver function tests frequently return to normal before rising again. Test results obtained over a period of adequate length must therefore be evaluated by the physician before the donor can be readmitted to the programme.

4.5.3 *Tests for plasma donors*

The following tests shall be performed at each donation:

- Measurement of haemoglobin concentration or erythrocyte volume fraction.
- Determination of total serum protein concentration, which shall be at least 60 g/l.
- An approved test for HBsAg, which shall be negative.
- An approved test for anti-HIV, which shall be negative.
- An approved test for anti-HCV, which shall be negative.

The following tests shall be performed initially and then every four months or after every 10 donations, whichever time interval is longer:

- If required by the national control authority, a serological test for syphilis, which shall be negative.
- Urine analysis for glucose and protein, which shall be negative.
- Serum protein electrophoresis: this shall be normal (unusual changes in a donor's results may be more significant than absolute values). The albumin and globulin concentrations may be calculated from the known total protein value, and shall be: albumin, minimum 35 g/l; IgM, minimum 0.5 g/l; IgG, between 5 and 20 g/l.
- Liver function tests.

When determination of serum alanine aminotransferase is required, the enzyme concentration measured photometrically using approved reagents shall be no more than two standard deviations above an established normal mean.

4.6 *Donors for platelet and leukocyte apheresis*

In general, platelet and leukocyte donors shall meet the general criteria for donors and the specific criteria for plasma donors (sections 4.1–4.5). In addition, platelet donors should not have taken aspirin or other platelet-active drugs for at least 72 h before donation.

The requirements to be satisfied in the performance of plateletpheresis and leukapheresis in order to ensure that there is no danger to donors and that the products obtained are of satisfactory quality are under active investigation in many countries. The following recommendations may be useful as guidance.

On the day of each donation, donors for plateletpheresis should have an absolute platelet number concentration ("count") of not less than 200×10^9/l and donors for leukapheresis should have an absolute granulocyte number

concentration of not less than 3×10^9/l. Both types of donor should have a normal differential leukocyte count and haemoglobin level.

Although levels of circulating platelets and leukocytes recover promptly in donors, data are not at present available from which the maximum numbers of platelets and leukocytes that can be safely collected from donors can be defined. The long-term effects of the repeated removal of cellular elements are not known.

Leukapheresis may entail the administration of drugs to donors and their exposure to colloidal agents to enhance the yield of granulocytes. Appropriate precautions should be taken to protect donors, such as investigation for latent diabetes by means of a glucose tolerance test if a donor is to be given corticosteroids.

Leukapheresis should be performed as part of the treatment of a patient with chronic myeloid leukaemia only if approved by the patient's attending physician. It is inadvisable to use the leukocytes from such patients.

4.7 *Donor immunization and plasma for special purposes*

4.7.1 *Plasmapheresis in donors with naturally acquired antibodies and other types of medically useful plasma*

Plasma may be collected by plasmapheresis from donors who have acquired immunity through natural infection or through active immunization with approved vaccines for their own protection, and from donors with plasma useful for diagnostic purposes as a result of acquired or congenital underlying conditions.

Donors with medically useful plasma may be identified by screening whole blood donations and by examining patients convalescing from specific diseases or vaccinated individuals, e.g. veterinary students who have received rabies vaccine or military recruits who have been immunized with tetanus toxoid. Unnecessary immunizations can be avoided by this approach.

The following are examples of medically useful plasma:

- Antibody-rich plasma for control reagents in diagnostic tests, such as those for anti-HIV, hepatitis A and B, cytomegalovirus, rubella, measles and uncommon infectious agents; plasma should be collected in appropriately isolated premises when products are being prepared that are known to be capable of transmitting infection.
- Plasma containing antibodies to human cellular and serum antigens of diagnostic use, for example in HLA (human leukocyte antigen) typing reagents, erythrocyte typing reagents and immunoglobulin allotyping reagents.
- Plasma containing reagents useful for diagnostic tests, such as reagin, rheumatoid factors, heterophile antibody and C-reactive protein.
- Factor-deficient plasma for specific assays, such as factor-VIII-deficient plasma. Donors who have received factor VIII are at increased risk of transmitting hepatitis B, hepatitis C and HIV; their plasma should therefore be collected in appropriately isolated premises.

4.7.2 *Precautions to be taken when handling blood or blood products containing infectious agents*

All blood and plasma may contain unknown infectious agents and must be handled accordingly. In addition, special precautions must be taken when handling infected donors and blood products known to contain infectious agents. The precautions to be taken might include:

- isolation by means of the appropriate timing or location of the procedures, special labelling and quarantine of the products collected, use of protective packaging with double wrapping in impervious plastic;
- disinfection of all work surfaces and equipment with a disinfectant of known efficacy, such as freshly prepared 0.25% sodium hypochlorite solution;
- protection of staff by means of adequate training, avoidance of aerosols and use of gloves, gowns, masks and eye protection; it is strongly recommended that such staff also be protected by immunization with hepatitis B vaccine;
- fulfilment of the labelling, shipping and waste-disposal requirements appropriate to the etiological agents in question.

4.7.3 *Immunization of donors*

There is a clinically valid need for specific immunoglobulins and plasma for therapeutic, prophylactic and diagnostic uses. Deliberate immunization of healthy volunteers may be necessary in addition to collection of plasma from convalescent patients and donors selected by screening for high levels of specific antibodies. The immunization of donors requires informed consent in writing and shall take into consideration all the requirements of the previous sections.

Donors shall be immunized with antigens only when sufficient supplies of material of suitable quality cannot be obtained from other appropriate donors, from donations selected by screening, or in the form of safe and efficacious licensed monoclonal antibodies. Donors must be fully informed of the risk of any proposed immunization procedure, and pressure shall not be brought to bear on a donor to agree to immunization. Women capable of child-bearing shall not be immunized with erythrocytes or other antigens that may produce antibodies harmful to the fetus. Donors of blood and those undergoing plasmapheresis shall, if necessary, undergo investigations that can reveal hypersensitivity to a proposed antigen (see also Part B, section 6).

An approved schedule of immunization shall be used. Every effort shall be made to use the minimum dose of antigen and number of injections. In any immunization programme, the following shall be taken into consideration as a minimum: (a) the antibody assay; (b) the minimum level of antibody required; (c) data showing that the dose, the intervals between injections and the total dosage proposed for each antigen are appropriate; and (d) the criteria for considering a prospective donor a non-responder for a given antigen. No donor shall be hyperimmunized with more than one

immunizing preparation unless the safety of the multiple procedure is demonstrated.

Potential donors should be:

- informed by a licensed physician of the procedures, risks and possible sequelae and how to report any adverse effects, and encouraged to take part in a free discussion (which, in some countries, is achieved in small groups of potential donors);
- encouraged to seek advice from their family doctor before agreeing to immunization;
- informed that any licensed physician of their choice will be sent all the information about the proposed immunization procedure;
- informed that they are free to withdraw consent at any time.

All vaccines used for immunizing donors shall be registered or recognized by the national health authority, but may be administered at doses and with schedules differing from those recommended for routine prophylactic immunization. Erythrocyte and other cellular antigens shall be obtained from an establishment approved by the national control authority.

Donors shall be observed for approximately 30 min following any immunization in order to determine whether an adverse reaction has taken place. Because reactions often occur 2–3 h after immunization, donors shall be advised of this possibility and instructed to contact the facility's physician if a reaction is suspected in the first 12 h after immunization. Reactions may be local or systemic. Local reactions, which may be immediate or delayed, take the form of redness, swelling or pain at the injection site. Systemic reactions may include fever, chills, malaise, arthralgia, anorexia, shortness of breath and wheezing.

4.7.4 *Immunization with human erythrocytes*

Erythrocyte donors. A donor of erythrocytes for the purposes of immunization shall meet all the general health criteria for donors (see sections 4.3 and 4.4). In addition, the donor shall not have had a blood transfusion at any time.

The volume of erythrocytes drawn from a donor should not exceed 450–500 ml of whole blood in any eight-week period.

At each donation the donor shall be found to be negative for syphilis, HBsAg, anti-HIV, antibody to hepatitis B core antigen (anti-HBc), anti-HCV and antibodies to human T-cell lymphotropic viruses (anti-HTLV). The serum level of aminotransferases should be within normal limits as established by the national control authority.

Erythrocyte phenotyping shall be done for ABO as well as for C, D, E, c, e, Kell and Fy^a. Phenotyping for other specificities is often desirable and is recommended especially for Jk^a, Jk^b, Fy^b, S and s.

Ideally erythrocytes obtained for immunization purposes should be frozen for at least 12 months before use and the donor should be recalled and retested for anti-HIV, anti-HCV, anti-HBc, HBsAg and anti-HTLV before the stored cells are used for immunization.

Where suitable facilities for freezing erythrocytes are not available, national control authorities may authorize the use of cells from a single donor to immunize no more than three persons (preferably who have not previously had a blood transfusion) in an initial 12-month period, during which monthly determinations of anti-HIV, anti-HCV, anti-HBc, HBsAg and serum alanine aminotransferase should be made in both the donor and the recipients. If, after 12 months, the initial three recipients show no clinical or laboratory evidence of hepatitis, HIV infection or other blood-transmissible diseases, the donor may be considered acceptable for providing erythrocytes for immunization. As small a number of donors of erythrocytes should be used as possible.

Collection and storage of erythrocytes. Erythrocytes shall be collected under aseptic conditions into sterile, pyrogen-free containers in an appropriate proportion of an approved anticoagulant. They may then be dispensed in aliquots under aseptic conditions into single-dose, sterile, pyrogen-free containers for storage. The microbiological safety of the dispensing environment shall be validated.

Erythrocytes should be stored frozen for at least 12 months to permit retesting of donors for disease markers. The method selected should have been validated such that there is 70% cell recovery *in vivo*. Erythrocytes should be washed after storage to remove the cryoprotective agent.

Adequate sterility data to support the requested shelf-life for stored erythrocytes should be submitted by the manufacturer to the national control authority. A test for bacterial and fungal contamination should be made on all blood dispensed in aliquots in an open system (*9*). The test should also be performed on at least one single-dose vial from each lot of whole blood that has been stored unfrozen for more than seven days. The test should be made on the eighth day after collection and again on the expiry date. Cultures for the sterility test should be maintained for at least 14 days, with subculturing on day 3, 4 or 5.

Erythrocyte recipients. The following additional testing of erythrocyte recipients is necessary:

- The recipient should be phenotyped for ABO, Rh, Kell and Duffy antigens before immunization. Kell-negative and/or Fy(a–) persons should not receive Kell-positive or Fy(a+) cells except for the specific purpose of producing anti-Kell or anti-Fy^a. Only ABO-compatible erythrocytes may be transfused. Matching of Jk^a, Jk^b, Fy^b, S and s phenotypes is also desirable.
- Screening for unexpected antibodies by methods that demonstrate coating and haemolytic antibodies should include the antiglobulin method or a procedure of equivalent sensitivity.

Prospective erythrocyte recipients in whom antibody screening tests demonstrate the presence of erythrocyte antibodies (other than those deliberately stimulated through immunization by the plasmapheresis centre) should be asked whether they have ever been pregnant or had a

transfusion, a tissue graft or an injection of erythrocytes for any reason. This history should form part of the permanent record and should identify the cause of immunization as clearly as possible. Recipients should be notified in writing of any specific antibodies developed after injection of erythrocytes. The national control authority should be notified annually in writing of unexpected antibodies induced by immunization, and the immunized donor should carry a card specifying the antibodies.

Immunization schedules. Erythrocytes used for immunization purposes shall not be administered as part of any plasmapheresis procedure. Such immunization may be performed on the same day as plasmapheresis, but only after it and as a separate procedure.

To minimize the risk of infection to the donor, the immunization schedule should involve as few doses of erythrocytes as possible.

For primary immunization two injections of erythrocytes, each of about 1–2 ml and given three months apart, elicit antibody formation within three months of the second injection in approximately 50% of volunteers; the result is not improved by injecting larger amounts or giving more frequent injections.

It is advantageous to choose as donors of anti-D (anti-Rh_0) volunteers who are already immunized, since useful levels of anti-D are then usually attained within a few weeks of reimmunization. In some people, the level of antibody reaches its maximum within the first three weeks and will not increase after further immunization. In others, antibody levels may continue to rise for more than 12 months when injections of 0.5–1 ml of erythrocytes are given at intervals of five to eight weeks. About 70% of immunized volunteers eventually produce antibody levels well above 100 IU/ml. Once attained, such levels can be maintained by injections of 0.1–0.5 ml of erythrocytes at intervals of two to nine months, as required. If injections of erythrocytes are discontinued, antibody levels usually fall appreciably within 6–12 months.

The baseline antibody titre of every recipient of erythrocytes should be established, and the antibody response, including both type and titre, should be monitored monthly.

Erythrocytes to be used for immunization purposes should be selected, for each recipient, by a licensed physician.

Risks to recipients. Recipients of erythrocytes for immunization purposes may run the risk of:

- viral hepatitis (B and C) and HIV infection;
- other infectious diseases;
- HLA immunization;
- the production of unwanted erythrocyte antibodies that may complicate any future blood transfusion;
- a febrile reaction if the antigen dose is too great;

– the production of antibodies that may interfere with future organ transplantation if it is needed.

Record-keeping. Records of erythrocyte donors and of the recipients of their erythrocytes should be maintained and cross-referenced.

5. Collection of blood and plasma

A number of precautions must be taken in the collection of blood and plasma, as described in the following sections.

5.1 *Blood collection and apheresis procedures*

The skin of the donor at the site of venepuncture shall be prepared by a method that has been shown to give reasonable assurance that the blood collected will be sterile. Blood shall be collected into a container by means of an aseptic method. The equipment for collecting the sterile blood may be closed or vented provided that the vent is designed to protect the blood against microbial contamination.

With apheresis procedures, care shall be taken to ensure that the maximum volume of erythrocytes is returned to the donor by intravenous infusion. If the red cells cannot be returned to the donor, no further collection should be made until the donor has been re-evaluated. Several checks shall be made to ensure that donors receive their own erythrocytes, including identification of the containers of erythrocytes by donors before re-infusion. Haemolytic transfusion reactions are avoidable, since they are caused by the accidental infusion of incompatible erythrocytes. Personnel involved in reinfusion procedures should be adequately trained to prevent them. The signs and symptoms are hypotension, shortness of breath, stomach and/or flank pain, apprehension, cyanosis and haemoglobinuria.

If a haemolytic transfusion reaction occurs, the infusion of cells to all donors at the centre concerned should be discontinued until the identity of all containers of erythrocytes has been checked. Automated plasmapheresis is preferred to manual plasmapheresis in some institutions because of its greater safety.

5.1.1 *Summary of minimum general requirements for apheresis*

Equipment. This must be electrically safe and non-destructive for blood elements; disposable tubing must be used wherever there is blood contact. In addition, equipment must be accessible to detailed inspection and servicing and its decommissioning should not significantly interrupt the programme. It should also be provided with suitable automatic alarms.

Procedure. This must be non-destructive for blood elements and aseptic; there must be adequate safeguards against air embolism.

Disposables. These must be pyrogen-free, sterile and biocompatible (e.g. there must be no activation of enzyme systems).

5.1.2 *Adverse reactions*

Provision must be made to prevent and treat any adverse reactions in donors. As with any medical procedure involving the treatment of individuals, adverse reactions may occur with blood collection and plasmapheresis. Almost all such reactions are mild and transient, but an occasional serious reaction may occur. The possibility of adverse reactions, though remote, should be anticipated and adequate provision should be made to ensure that care is available to donors. Initial and continuing training in emergency care is mandatory for personnel. If any serious adverse reaction occurs, a physician should be called.

5.1.3 *Types of adverse reaction*

Vasovagal syncope. This is most likely to occur with new donors. The signs and symptoms are hypotension, bradycardia, syncope, sweating and (rarely) convulsions.

Local infection, inflammation and haematoma at the phlebotomy site. Reactions of this type are best prevented by adequate preparation of the venepuncture site and by training phlebotomists in proper methods of initiating blood flow. The symptoms are localized pain and redness and swelling at the phlebotomy site.

Allergic and anaphylactoid reactions. These may occur during the introduction of saline into the donor while red cells are being processed, or during reinfusion of red cells. The signs and symptoms are urticaria, burning in the throat, tightness of the chest, wheezing, pain in the abdomen and hypotension.

Systemic infection. Care should be taken at all stages of plasmapheresis to avoid the transmission of infectious organisms to the donor.

5.2 Containers

The original blood container or a satellite attached in an integral manner shall be the final container for whole blood and red cells, with the exception of modified red cells, for which the storage period after processing should be as short as possible and certainly not longer than 24 h. Containers shall be uncoloured and translucent and the labelling shall be placed in such a position as to allow visual inspection of the contents. They shall be sterilized and hermetically sealed by means of suitable closures so that contamination of the contents is prevented. The container material shall not interact adversely with the contents under the prescribed conditions of storage and use.

The specifications for containers should be approved by the national control authority (*10, 11*).

If sterile docking devices are not available, closed blood-collection and processing systems should be used to prepare blood components.

5.3 *Anticoagulants*

The anticoagulant solution shall be sterile, pyrogen-free and of a composition such as to ensure that the whole blood and separate blood components are of satisfactory safety and efficacy.

Commonly used anticoagulant solutions are acid–citrate–glucose, citrate–phosphate–glucose and citrate–phosphate–glucose–adenine; the amount of adenine used varies in different countries. Solutions of adenine, glucose and mannitol used for red cell preservation may be added after removal of the plasma.

For plasmapheresis, sodium citrate as a 40 g/l solution is widely used as an anticoagulant.

5.4 *Pilot samples*

Pilot samples are blood samples provided with each unit of whole blood or of red blood cells. They shall be collected at the time of donation by the person who collects the whole blood. The containers for pilot samples shall be marked at the collection site before the samples are collected, and the marking used must be such that the sample can be identified with the corresponding unit of whole blood. Pilot samples must be collected by a technique that does not compromise the sterility of the blood product.

Pilot samples should be attached to the final container in a manner such that it will later be clear whether they have been removed and reattached.

5.5 *Identification of samples*

Each container of blood, blood components and pilot and laboratory samples shall be identified by a unique number or symbol so that it can be traced back to the donor and from the donor to the recipient. The identity of each donor shall be established both when donor fitness is determined and at the time of blood collection.

When blood-derived materials are transferred to a fractionation plant, the following details shall be provided by the supplier:

- name and address of collecting centre,
- type of material,
- donor identification,
- date of collection,
- results of mandatory tests,
- conditions of storage,
- other details required by the fractionator,
- name and signature of responsible person,
- date.

Part B. Requirements for single-donor and small-pool products

6. General considerations

These requirements for single-donor and small-pool products cover the methods used to prepare products directly from units of whole blood or of components collected by apheresis, starting with the testing of the units and proceeding to the separation of the various cell and plasma protein components. Among the products that may be prepared in small pools (12 donors or fewer) are cryoprecipitated factor VIII and platelets. In addition to tests on the units of whole blood that provide information on the safety, efficacy and labelling of the components, specific tests are included, where applicable, to ensure the quality of various components.

It is important to note that single-donor and small-pool products have certain specialized uses other than therapeutic application to correct deficits in patients. Although not dealt with further in these Requirements, these uses include the stimulation of plasma donors with red blood cells in order to raise antibody levels for the preparation of anti-D (anti-Rh_o) immunoglobulin (*12*) and special blood-grouping reagents. It is of the utmost importance that the donors of cells and plasma for such purposes be carefully studied both initially and on a continuing basis to minimize the likelihood of the transmission of infectious diseases to recipients. The use of red cells, stored frozen, that have been demonstrated to be free from infectious agents by retesting the donor 12 months after the initial collection reduces the risk of such transmission to volunteers for immunization.

Plasma donors may also be immunized with viral or bacterial antigens for the preparation of specific immunoglobulin products. All donor immunization procedures must be planned and carried out under the supervision of a physician who is familiar with the antigens being used and especially with the reactions or complications that may occur. Donors being immunized shall have been fully informed of all known hazards and shall have given their written informed consent to the procedures.

Donor immunization practices are considered in more detail in Part A, section 4.7.

Minimum general requirements for apheresis are summarized in Part A, section 5.1.1.

7. Production and control

7.1 *General requirements*

Single-donor and small-pool products shall comply with any specifications established by the national control authority. Cellular blood components and certain plasma components may deteriorate during separation

or storage. Whatever the method of separation (sedimentation, centrifugation, washing or filtration) used for the preparation of cell components, therefore, it is important that a portion of plasma protein sufficient to ensure optimum cell preservation be left with the cells, except when a cryoprotective substance is added to enable them to be stored for long periods in the frozen state, or additive solutions (for example containing adenine, glucose and mannitol) are used for the same purpose for liquid storage.

The methods employed for component separation should be checked before they are introduced. The characteristics assessed might include yield of the component, purity, *in vivo* recovery, biological half-life, functional behaviour and sterility.

The nature and number of such checks should be determined by the national control authority.

Immediately before issue for transfusion or for other purposes, blood components shall be inspected visually. They shall not be issued for transfusion if abnormalities of colour are observed or if there is any other indication of microbial contamination or of defects in the container.

Blood components shall be stored and transported at the appropriate temperature. Refrigerator or freezer compartments in which components are stored shall contain only whole blood and blood components. Reagents required for use in testing may be stored in a separate section of the same refrigerator or freezer provided that they have been properly isolated and are in suitable containers.

7.2 Testing of whole blood and plasma

7.2.1 *Sterility*

Each donation of whole blood intended for transfusion and each preparation of component cells constitutes a single batch. Single batches shall not be tested for sterility by any method that entails breaching the final container before the blood is transfused.

The national control authority may require tests for sterility to be carried out at regular intervals on final containers chosen at random and at the end of the storage period. The purpose of such tests is to check on the aseptic technique used for taking and processing the blood and on the conditions of storage.

7.2.2 *Laboratory tests*

Laboratory tests shall be made on laboratory samples taken either at the time of collection or from the pilot samples accompanying the final container, labelled as required in Part A, section 5.

In some countries, test reagents, in particular those used for blood-grouping and for detecting anti-HIV, anti-HCV and HBsAg, must be approved by the national control authority.

The results of the tests shall be used for ensuring the safety and proper labelling of all components prepared from units of whole blood.

7.2.3 *Tests for infectious agents*

Syphilis. Each donation of whole blood shall, if required by the national control authority, be subjected to a serological test for syphilis. If so tested, only units giving negative results shall be used for transfusion or component preparation.

Viral hepatitis. Each unit of blood or plasma collected shall be tested for HBsAg and anti-HCV by a method approved by the national control authority and only those giving a negative result shall be used (*13*). Units giving a positive result shall be so marked, segregated and disposed of by a method approved by the national control authority, unless designated for the production of a reagent or experimental vaccine in an area designed and segregated for such production.

In some countries plasma pools are also tested.

The label on the container or the record accompanying the container should indicate the geographical source of the blood or plasma as well as whether and how the material has been tested for HBsAg and anti-HCV.

Liver function tests, such as serum transaminase determinations, are used in some countries to detect liver damage that may be associated with hepatitis.

Anti-HIV-1 and anti-HIV-2. Blood for transfusion and for use in the preparation of blood components must be tested by a method approved by the national control authority for antibodies to HIV-1 and HIV-2 and be found negative. However, when other important factors outweigh the benefits of such testing (e.g. in emergencies) formal arrangements, approved in advance by the national control authority, should be in place that enable the prescribing physician to have access to an untested product. In all such cases, retrospective testing of the pilot sample shall be performed.

Other infectious agents. It is important for the national control authority to reassess testing requirements from time to time in the light of current knowledge, the prevalence of infectious agents in different populations and the availability of tests for serological markers of infection. For example, human retroviruses other than HIV have been described (HTLV types 1 and 2) and more may be identified in the future.

7.3 **Blood-grouping**

Each unit of blood collected shall be classified according to its ABO blood group by testing the red blood cells with anti-A and anti-B sera and by testing the serum or plasma with pooled known group A (or single subtype A_1) cells and known group B cells. The unit shall not be labelled as to ABO group unless the results of the two tests (cell and serum grouping) are in agreement. Where discrepancies are found in the testing or the donor's records, they shall be resolved before the units are labelled.

In countries where polymorphism for the D (Rh_o) antigen is present, each unit of blood shall be classified according to Rh blood type on the basis of

the results of testing for the D (Rh_o) red cell antigen. The D (Rh_o) type shall be determined with anti-D (anti-Rh_o) reagents.

With the high-strength antisera and sensitive techniques now available, it is usually considered unnecessary to use the D^u test if the cells are found to be D-negative in routine testing.

7.4 Red cells

Whole blood for the preparation of all components shall be collected as described in Part A, section 5, and tested as described in Part B, section 7.2.

Red cells shall be processed under aseptic conditions and whenever possible in a closed system. The sterility of all components shall be maintained during processing by the use of aseptic techniques and sterile pyrogen-free equipment. The methods shall be approved by the national control authority, and a written description of the procedures shall be prepared for each product, covering each step in production and testing. Proposals for any procedural modifications shall be submitted to the national control authority for approval before they are implemented.

The following may be prepared for therapeutic purposes (see pages 40–41 for definitions):

- red cells;
- red cells suspended in additive solution;
- modified red cells:
  - red cells, leukocyte-depleted;
  - red cells, leukocyte-poor;
  - red cells, washed;
  - red cells, frozen;
  - red cells, deglycerolized.

7.4.1 *Methods and timing of separation*

Red cells shall be prepared from whole blood collected in plastic bags or in glass bottles.

Multiple-plastic-bag systems with sterile docking devices are preferable because they minimize the risk of microbial contamination by providing completely closed systems. They are easy to handle and are disposable. The use of glass bottles is cheaper but has the disadvantage that the system is then an open or vented one, so that separation must be carried out under strictly aseptic conditions in sterile rooms or laminar-flow cabinets and microbiological monitoring is necessary. The same conditions also apply to the separation procedure when plasma is transferred from disposable single plastic bags to separate containers.

All surfaces that come into contact with the blood cells shall be sterile, biocompatible and pyrogen-free. If an open plastic-bag system is used, i.e. the transfer container is not integrally attached to the blood container and the blood container is opened after blood collection, the plasma shall be separated from the cells under conditions such that the original container is kept under positive pressure until it has been sealed. If the separation

procedure involves a vented system, i.e. if an airway is inserted into the container for withdrawal of the plasma, the airway and vent shall be sterile and constructed so as to exclude microorganisms.

In some countries, the sterility of products prepared in open systems is monitored by testing a sample of at least 2% of the units. The national control authority should approve the system used.

The final containers for red cells (but not necessarily modified red cells) shall be the containers in which the blood was originally collected or satellite containers attached in an integral manner. If pilot samples are detached from the blood container during removal of any component, such samples shall be reattached to the container of red cells. The removal and reattachment of the pilot samples shall be recorded conspicuously (with a signature) on the label of the unit. The final containers for all other components shall meet the requirements for blood containers given in Part A, section 5.2. If the final container differs from the container in which the blood was originally collected, it shall be given a number or other symbol to identify the donor(s) of the source blood. Whenever appropriate, the secondary final container shall be similarly labelled while attached to the primary final container.

The timing and the method of separation (centrifugation, undisturbed sedimentation or a combination of the two) will depend on the components to be prepared from the donation. When platelets and coagulation factors are being prepared from the same donation, the components shall be separated as soon as possible after withdrawal of the blood from the donor.

Separation should preferably be effected within 8 h of blood donation.

When platelets and coagulation factors are to be prepared, it is especially important that the venepuncture be performed in such a way as to cause minimal tissue damage so as to prevent the initiation of coagulation. The blood should flow freely without interruption and as rapidly as possible, and be mixed thoroughly with the anticoagulant.

If platelets are to be prepared from a unit of whole blood, the blood shall be kept at a temperature of 20–24 °C for up to 8 h until the platelet-rich plasma has been separated from the red blood cells.

Red cells may be prepared either by centrifugation or by undisturbed sedimentation before the expiry date of the original whole blood. Blood cells shall be separated by centrifugation in a manner that will not increase the temperature of the blood.

Sedimentation is the least expensive method for separation of red blood cells and does not require special equipment.

Repeated washing with saline and centrifugation and filtration are used to reduce the number of leukocytes and platelets and the volume of trapped plasma in red cells. Frozen red cells after thawing are also repeatedly washed with special solutions to remove cryoprotective agents while also preventing haemolysis.

7.4.2 *Expiry date*

The expiry date of whole blood and red cells prepared in a closed system from blood collected in acid–citrate–glucose or citrate–phosphate–glucose is generally 21 days after collection. The time of removal of plasma is not relevant to the expiry date of the red cells when the integrity of the container is not compromised.

The shelf-life of stored blood has been extended to 35 days by collecting the blood in acid–citrate–glucose supplemented with 0.5 mmol/l adenine or in a mixture of 0.5 mmol/l adenine and 0.25 mmol/l guanosine with extra glucose, and to 42 days by adding a solution containing adenine, glucose and mannitol. Recent studies indicate that it may also be possible to extend the shelf-life of stored blood to 35 days by collecting it in citrate–phosphate–glucose supplemented with 0.25 mmol/l adenine and extra glucose.

When red cells are prepared with very high erythrocyte volume fractions, an expiry date 14 days after collection is recommended in some countries because the cells may become glucose-deficient after this time. The erythrocyte volume fraction of red cells collected in citrate–phosphate–glucose–adenine should not exceed 0.9 if the expiry date is more than 21 days after collection.

The usefulness of acid–citrate–glucose is limited by the significant reduction in cell viability when the volume of cells collected is small, which is unavoidable for some donations.

Provided that sterility is maintained, the shelf-life of red cells is not influenced by the method of separation used. However, if an open system is used that does not maintain sterility, the expiry date shall be 24 h after separation and the cells should be used as soon as possible. Red cells and whole blood should be stored at 5 ± 3 °C and transported with wet ice in insulated boxes at 5 ± 3 °C. Care should be taken not to place containers directly on ice.

Refrigerated whole blood and red cells will warm up rapidly when placed at room temperature. Every effort should be made to limit the periods during which the products are handled at ambient temperatures in order to prevent the temperature from rising above 10 °C until they are used.

7.4.3 *Modified red cells*

Red cells, leukocyte-depleted and red cells, leukocyte-poor.

Because of the possibility of reactions, some countries require that red cells contain less than 2% of the leukocytes of the original whole blood.

Leukocyte depletion may be achieved by buffy-coat removal, freezing and washing, or by washing alone.

Leukocyte-poor red-cell concentrates are prepared by filtration.

Red cells, washed. Red cells can be washed by means of interrupted or continuous-flow centrifugation. If the first of these methods is used, the washing procedure shall be repeated three times.

Centrifugation should be carried out in refrigerated centrifuges. If such

equipment is not available, the washing solution should have a temperature of 5±3 °C.

Red cells can also be washed by means of reversible agglomeration and sedimentation using sugar solutions.

Washed red cells should be transfused as soon as possible and in any case not later than 24 h after processing if prepared in an open system that does not maintain sterility, unless the national control authority has specified a longer shelf-life. They should be stored at all times at 5±3 °C.

Requirements for pilot samples, labels and storage and transport temperatures are the same as those for unmodified red cells.

Red cells, frozen and red cells, deglycerolized. Red cells less than six days old are usually selected for freezing in order to minimize loss of yield due to haemolysis during processing.

Frozen red cells are red cells that have been stored continuously at low temperatures (−65 °C or below) in the presence of a cryoprotective agent. The red cells must be washed to remove the cryoprotective agent before use for transfusion. The methods of preparation, storage, thawing and washing used should be such as to ensure that at least 70% of the transfused cells are viable 24 h after transfusion. Storage at temperatures below −65 °C is usually necessary to achieve 70% recovery.

The cryoprotective agent in most common use is glycerol. The temperature of storage should be between −65 °C and −160 °C, depending on the glycerol concentration used.

The shelf-life of frozen cells below −65 °C is at least three years and may be much longer under certain circumstances, but the reconstituted (thawed and washed) red cells should be used as soon as possible and not later than 24 h after thawing unless a closed system is used.

Frozen cells are usually shipped in solid carbon dioxide ("dry ice") or liquid nitrogen, depending upon the glycerol concentration used. Deglycerolized red cells should be stored at a temperature of 1–6 °C and shipped at 5±3 °C.

Requirements for pilot samples and labels are the same as those for unmodified red cells.

7.5 Plasma

Single-donor plasma shall be obtained by plasmapheresis or from units of whole blood that comply with the requirements of Part A, section 5, and Part B, section 7.2.

Fresh-frozen plasma and frozen plasma should be stored in carefully monitored freezers equipped with recording thermometers and audio and visual alarms to give warning of mechanical or electrical failure. If refrigeration is interrupted for longer than 72 h and the temperature rises above −5 °C, the product may no longer be considered as fresh-frozen plasma, although testing may indicate that reasonable amounts of factor

VIII remain if the plasma has not become liquid. Repeated thawing and freezing may cause denaturation of plasma constituents and cause prekallikrein activation.

7.5.1 *Plasma, fresh-frozen*

Fresh-frozen plasma shall be prepared by separating plasma from whole blood and freezing it rapidly within 8 h of collection.

Ideally, fresh-frozen plasma should be prepared by rapid freezing using a combination of solid carbon dioxide and an organic solvent such as ethanol. If this procedure is used, it should have been shown that the container cannot be penetrated by the solvent or substances leached from the container into the contents. Fresh-frozen plasma should be stored at or below −20 °C, and below −30 °C if to be used for transfusion purposes.

Before use for infusion, fresh-frozen plasma should be thawed rapidly at 30–37 °C. Agitation of the container and/or circulation of water at a temperature of 37 °C during the thaw cycle will speed thawing. Once thawed, fresh-frozen plasma must not be refrozen. It can be stored at ambient temperature and should be used within 2 h of completion of thawing.

Fresh-frozen plasma shall have an expiry date one year from the date of collection.

Before its expiry date, fresh-frozen plasma may be used for preparing cryoprecipitated factor VIII. It may be used for the preparation of other pooled plasma fractions (e.g. factors I, II, VII, VIII, IX and X) at any time, even after its expiry date.

7.5.2 *Plasma, frozen*

Frozen plasma is, by definition, a plasma separated from whole blood more than 8 h after the latter has been collected, but the delay should be as short as possible. Frozen plasma may be used directly for transfusion or fractionation, or it may be freeze-dried as single-donor units. Plasma may be combined in small pools before freezing if it is to be used to prepare freeze-dried plasma.

The national control authority should determine the specific requirements for frozen plasma.

If frozen or freeze-dried plasma is intended to be used directly in patients without further processing, the blood shall be collected in such a manner and in containers of such a type as to allow aseptic handling, e.g. by means of closed systems.

In some countries, frozen plasma is given an expiry date five years from the date of collection.

Whenever the container of frozen plasma is opened in an open procedure, the method of handling shall avoid microbial contamination; as an additional precaution, sterile rooms or laminar-flow cabinets can be used. Delay in processing shall be avoided, and the ambient conditions shall be regulated so as to minimize the risk of contamination.

Plasma may be pooled at any time after collection.

7.5.3 *Plasma, freeze-dried*

Freeze-dried plasma shall be made from single units or small pools of fresh-frozen plasma or frozen plasma.

The storage conditions and expiry dates of different forms of freeze-dried plasma shall be approved by the national control authority. The product normally has a shelf-life of five years when stored at 5 ± 3 °C, but this will depend on the source material, storage conditions and residual moisture in the product. Pooled freeze-dried plasma has a significant potential for the transmission of infectious diseases. This is likely to be substantially diminished by the introduction of viral inactivation procedures applicable to plasma.

7.5.4 *Plasma, recovered*

Recovered plasma intended to be pooled for fractionation shall not be used directly for transfusion; a preservative shall not be added.

Plasma may be separated from whole blood at any time up to five days after the expiry date of the blood. The method used for separation shall avoid microbial contamination. As an additional precaution, sterile rooms or laminar-flow cabinets can be used.

If the plasma has been pooled, it shall be stored and transported frozen at or below -20 °C.

7.5.5 *Plasma, platelet-rich*

Platelet-rich plasma is a preparation containing at least 70% of the platelets of the original whole blood.

The preparation shall be separated by centrifugation, preferably within 8 h of collection of the whole blood. The temperature and time of processing and storage shall be consistent with platelet survival and maintenance of function.

To achieve the desired haemostatic effect, platelet-rich plasma shall be transfused as soon as possible after collection, and not later than 72 h afterwards, unless stored at 22 ± 2 °C in containers approved for a longer storage period.

7.6 **Platelets**

Platelets shall be obtained by cytapheresis or from whole blood, buffy coat or platelet-rich plasma that complies with the requirements of Part A, section 5, and Part B, section 7.2. Aspirin ingestion within the previous three days precludes a donor from serving as a source of platelets.

Whole blood or platelet-rich plasma from which platelets are derived shall be maintained at 22 ± 2 °C until the platelets have been separated.

The separation shall preferably be performed within 8 h of collection of the whole blood. Blood shall be obtained from the donor by means of a single venepuncture giving an uninterrupted flow of blood with minimum damage to the tissue. It must have been demonstrated that the time and speed of centrifugation used to separate the platelets will produce a suspension without visible aggregation or haemolysis.

The national control authority shall determine the minimum acceptable number of platelets that should be present in the products prepared (e.g. 5.5×10^{10}).

A pH of 6.5–7.4 shall be maintained throughout storage of platelets. The volume of plasma used to resuspend platelets will be governed by the required pH of the platelet suspension at the end of its shelf-life, but shall be no less than 50 ± 10 ml.

Licensed artificial suspension media may be used to replace plasma.

Platelets stored at 5 °C are inferior to the same product stored at 22 ± 2 °C. Cold storage should be avoided where possible.

When stored at 22 ± 2 °C, platelet products shall be gently agitated throughout the storage period.

Platelet products with high platelet counts that are stored at 22 ± 2 °C may need to contain as much as 70 ml of plasma or more if the pH is to be maintained above 6.5 throughout the storage period. This period may be as long as seven days for containers made of certain special plastics, but it is prudent to restrict platelet storage to five days because of the risk of bacterial contaminants.

The product should be ABO typed and, in countries where D (Rh_o) is polymorphic, D (Rh_o) typed; it may also be desirable to know the HLA type.

The material of which the final container used for platelets is made shall not interact with the contents under normal conditions of storage in such a manner as to have an adverse effect on the product.

The requirements for labelling the final container are given in section 7.9. In addition to the customary data, the label shall bear: (a) the recommended storage temperature; (b) the statement that, when stored at 22 ± 2 °C, the platelets should be gently agitated throughout storage to obtain maximum haemostatic effectiveness; and (c) a statement to the effect that the contents should be used as soon as possible, and preferably within 4 h once the containers have been opened for pooling.

7.6.1 *Monitoring the quality of platelets*

Units randomly selected at the end of their shelf-life shall be tested on a regular basis. They shall be shown to have: (a) plasma volumes appropriate to the storage temperature; and (b) a pH between 6.5 and 7.4.

The number of units and of platelets to be tested shall be specified by the national control authority.

Some countries require there to be no visible contamination by red cells.

7.6.2 *Expiry date*

The expiry date of platelets processed in a closed system shall be 72 h after the original whole blood was collected, unless they are stored in a plastic container approved by the national control authority for a longer storage period.

Platelets prepared in an open system should be used within 4 h of preparation if stored at 22 ± 2 °C, unless the procedure used has been shown to allow a longer storage period.

Single-donor platelet concentrates may be pooled for one recipient under aseptic conditions before issue. Such small pools should be used as soon as possible, and within 4 h of preparation if stored at room temperature.

7.7 Leukocytes

Leukocytes are obtained by the separation of whole blood or by apheresis, and may contain a large number of platelets and red blood cells, depending on the method of preparation. When leukocytes are obtained from units of whole blood, such units shall comply with the requirements of Part A, section 5, and Part B, section 7.2.

The methods used to process leukocytes shall comply with the requirements and recommendations given in section 7.4.1 for the separation of red cells.

The label on the final container shall bear, in addition to customary data, instructions to use the leukocytes as soon as possible and in any case not more than 4 h after the container has been opened for pooling. The temperature of storage and transport shall be 22 ± 2 °C.

Leukocytes can be separated from blood by centrifugation, sedimentation or leukapheresis. To obtain a sufficient number, the leukocytes from units obtained from several healthy donors may have to be pooled.

Leukapheresis by continuous-flow filtration or centrifugation is the most efficient way of obtaining leukocytes, since it gives large numbers of high-quality cells from a single donor.

If centrifugation of whole blood is used, 30–60% of the leukocytes present in the original whole blood may be recovered.

Approximately 90% of the leukocytes present in the original whole blood can be separated by sedimentation of the red cells, accelerated by the addition of suitable substances with high relative molecular mass.

Leukocytes should be negative for cytomegalovirus.

The product should be ABO typed and, in countries where D (Rh_o) is polymorphic, D (Rh_o) typed; it may also be desirable to determine the HLA type. If not HLA typed, leukocytes should be irradiated.

The large number of red cells present in products prepared by some methods makes compatibility testing before transfusion necessary.

7.7.1 *Testing of leukocytes*

The number of units to be tested and the leukocyte yield (number) required shall be specified by the national control authority.

7.7.2 *Expiry date*

The expiry date of leukocytes shall be 24 h after collection of the original whole blood.

7.8 *Cryoprecipitated factor VIII*

Cryoprecipitated factor VIII is a crude preparation of factor VIII. It shall be obtained from single units or small pools of plasma derived either from units of whole blood that comply with the requirements of Part A, section 5, and Part B, section 7.2, or by plasmapheresis.

The product may be prepared as a pool from a small number of donations, usually four to six but not exceeding ten. It may be freeze-dried. However, preparations of cryoprecipitated factor VIII carry the risk of viral transmission unless they have undergone specific virucidal procedures during manufacture.

The method of thawing and harvesting the cryoprecipitate shall have been shown to yield a product containing an adequate activity of factor VIII (see section 7.8.1).

In procuring source material for coagulation factors, the following technical considerations should be borne in mind:

- In order to prevent coagulation, venepuncture should performed in such a way that tissue damage is minimal. The blood should flow freely without interruption, and be mixed thoroughly with anticoagulant during collection.
- Microbial contamination should be avoided during separation of the plasma by using multiple-plastic-bag closed systems or laminar-flow cabinets if an open procedure is used.
- The recovery of factor VIII depends on the interval between venepuncture and freezing of the plasma, the temperature at which the plasma is held and the freezing method. While a useful product may be obtained with plasma frozen as late as 18–24 h after phlebotomy, freezing the plasma as early and as rapidly as possible is strongly recommended.
- Ideally, fresh-frozen plasma should be prepared by rapid freezing using a combination of solid carbon dioxide and an organic solvent such as ethanol. Fresh-frozen plasma should be stored at or below -20 °C. Contamination of the plasma by the solvent or leaching of substances from the container into the plasma should be avoided.
- If the temperature of the thawed plasma exceeds 2 °C, a high proportion of the factor VIII is lost in the supernatant. During thawing or separation of the supernatant plasma, therefore, the temperature should not be allowed to exceed 2 °C. The plasma may be separated while there is still a small quantity of the ice present in the plasma

container. Increasing the speed of thawing by circulating air or water at a temperature of 0 °C is believed to increase the yield of factor VIII.

7.8.1 *Testing of cryoprecipitated factor VIII*

Randomly selected units shall be tested for potency and sterility on a regular basis. The number of units to be tested shall be specified by the national control authority. The freeze-dried preparation shall dissolve without any signs of precipitation in the solvent recommended by the manufacturer within 30 min when held at a temperature not exceeding 37 °C.

The potency of cryoprecipitated factor VIII shall be compared with that of an appropriate plasma or intermediate-purity standard, by measuring its ability to correct the prolonged activated partial thromboplastin time of haemophilia A plasma or by another suitable method.

When cryoprecipitated factor VIII is produced from fresh-frozen plasma (frozen within 8 h of donation), the yield should be greater than 400 IU/l of starting plasma. Plasma frozen after this time will yield less cryoprecipitated factor VIII.

In many laboratories, the average yield of factor VIII is 400 IU/l of starting plasma. The average yield of factor VIII as freeze-dried cryoprecipitate is then at least 300 IU/l of starting plasma. Whether this yield can be obtained elsewhere will depend on local technical possibilities. In some countries, the yields will be much lower, and the national control authority should decide as to the yield that is acceptable.

7.8.2 *Expiry date*

The frozen product shall be stored at or below -20 °C (if possible below -30 °C) and shall have an expiry date one year from the date of collection. The freeze-dried product shall be stored at 5 ± 3 °C and shall also have an expiry date one year from the date of collection. After thawing or reconstitution, cryoprecipitated factor VIII should be kept at 20–24 °C. It shall be used as soon as possible and in any case not more than 4 h after its container has been opened for pooling or reconstitution.

7.9 *Labelling*

After having been tested and before being issued for transfusion, units of single-donor and small-pool products shall be identified by means of container labels that clearly state at least the following information:

- the proper name of the product;
- the unique number or symbol identifying the donor(s);
- the expiry date, and when appropriate, the expiry time after reconstitution;
- any special storage conditions or handling precautions that are necessary;
- a reference to a package insert containing instructions for use, warnings and precautions;

- the name and address of the blood donor centre and, where applicable, the manufacturer and distributor;
- the average content in International Units of activity, where appropriate.

The results of red cell grouping shall be stated on the label of whole blood, red cells, fresh-frozen plasma (for clinical use), platelets and leukocytes but not necessarily on that of cryoprecipitated factor VIII.

Part C. Requirements for large-pool products

8. Introduction

A number of requirements common to albumin, plasma protein fraction, immunoglobulin preparations and coagulation-factor concentrates are given in Parts A and B, sections 3-7. However, for clarity, it has proved convenient to bring together in Part C certain specific requirements applicable to these products when manufactured on a large scale.

The source material for the large-scale preparation of blood products should comply with the relevant provisions of Parts A and B.

9. Buildings

The buildings used for the fractionation of plasma shall be of suitable size, construction and location to facilitate their proper operation, cleaning and maintenance in accordance with the requirements of Good Manufacturing Practices for Pharmaceutical (*7*) and Biological (*8*) Products. They shall comply with the Guidelines for National Authorities on Quality Assurance for Biological Products (*6*) and in addition provide adequate space, lighting and ventilation for the activities listed below.

Each of listed activities is an important integral part of the production procedure, and countries wishing to start manufacturing large-pool blood products and related substances should not do so unless adequate provision can be made for all of them.

9.1 *Storage of whole blood and plasma*

Whole human blood and plasma shall be stored frozen or refrigerated in separate facilities that are used only for this purpose. The source materials shall remain in quarantine until the results of testing show that they are suitable for introduction into the fractionation premises.

9.2 *Separation of cells and fractionation of plasma*

Cells shall be separated and plasma fractionated in a building isolated from those where non-human proteins or microbiological materials, such as vaccines, are manufactured or processed and separate from the animal house.

In some countries, cell constituents are separated in an area separate from that where plasma is fractionated.

9.3 *Supply and recovery of ancillary materials*

Adequate facilities shall be provided for the supply of ancillary materials, such as ethanol, water, salts and polyethylene glycol.

Facilities for the recovery of organic solvents used in fractionation may also be provided.

9.4 *Viral inactivation*

A separate area shall be provided for all processing subsequent to the completion of viral inactivation procedures when these are carried out at a stage in production before aseptic dispensing and filling (see section 9.5).

9.5 *Freeze-drying, filling, packaging, labelling and storage*

Separate facilities shall be used for the freeze-drying, filling, labelling and packaging of containers. A separate area shall be provided for the storage of labels, package inserts and packages. Another separate area shall be used for the storage of final containers before dispatch.

9.6 *Keeping of records*

Adequate provision shall be made for keeping records of all donors, materials, fractionation steps, quality-control procedures and results, of the distribution of the final products and of the disposal of potentially infectious materials. Records should be retained for at least two years beyond the expiry date of the products to which they relate.

Some manufacturers might wish to extend this period to cover any future legal disputes.

9.7 *Quality control*

Separate facilities shall be provided for quality control, including haematological, biochemical, physicochemical, microbiological, pyrogen and safety testing.

9.8 *Disposal of infective material*

Provision shall be made for the suitable disposal of potentially infectious materials by autoclaving or incineration according to good manufacturing practices.

The disposal of these materials should comply with local legislation.

10. Equipment

Equipment used for the collection, processing, storage and distribution of source materials and large-pool blood products shall comply with the requirements of Good Manufacturing Practices for Pharmaceutical (*7*) and Biological (*8*) Products.

Particular attention shall be paid to:

- The maintenance, monitoring and recording of the operation of continuously operating equipment, the validation of its reliability and the provision of stand-by equipment.
- The suitability and compatibility of the surfaces of all materials (e.g. filter medium, glass, stainless steel, plastic and rubber) that come into contact with the products.

  Metal surfaces that come into contact with proteins should be resistant to scratching. The surfaces of some materials can denature certain proteins or activate certain coagulation factors.

- The ease and efficiency with which equipment can be cleaned and, where necessary, sterilized. Any bactericidal agent used shall be capable of being completely eliminated before the equipment is used.

  Caution should be exercised in the use of detergents because of their possible effects on the final product; tests should be made to ensure that they do not have any adverse effect on it.

- The provision of suitable facilities for decontamination and for the disposal of potentially infective materials and equipment.

11. Provision of support services

A number of support services are essential for the fractionation of source materials.

11.1 *Water supply*

An adequate supply of suitable pyrogen-free water shall be provided for use during the fractionation process and for the reconstitution and/or dilution of the plasma fractions before filling and freeze-drying.

The two most commonly used types of water are pyrogen-free distilled water and pyrogen-free deionized water, each of which should be maintained at 80°C. Water preparation and delivery systems should be tested at regular intervals for endotoxin content and conductance. The water system should be a continuously circulating one and should have no dead ends.

Water for injections is generally used for the preparation of final products (*14*).

11.2 *Steam supply*

An adequate supply of steam shall be provided for the operation of sterilizing and cleaning equipment. The steam shall be clean and have the quality of water for injections.

11.3 *Other support facilities*

Other support facilities required are:

- A supply of electrical and thermal energy.

- A means of refrigeration for:
  - storing various source materials and fractions;
  - keeping the various fractionation areas at the correct temperature;
  - keeping the process equipment at the correct temperature;
  - storing final products under test;
  - storing final products awaiting dispatch.
- A system of ventilation providing the following two grades of filtered air:
  - air filtered to remove particles of 5 µm or greater in diameter, which shall be supplied to the entire work area; and
  - air passed through a filter with a retention capacity of more than 99.95% for particles greater than 0.5 µm in diameter, which shall be supplied at a positive pressure to areas where aseptic dispensing is to take place.

Other support facilities may include solvent recovery and a sewage disposal service. Sewage disposal must be carried out in accordance with the sanitary standards of the competent health authority.

Proteinaceous sewage from a plasma processing plant is highly nitrogenous and has a high biological oxygen demand; it should therefore not be discharged untreated.

These support facilities shall be located separately from the main process areas and in a place where the conditions (light, physical access, etc.) are conducive to the establishment of effective and routine preventive maintenance programmes. The equipment shall incorporate devices capable of monitoring and recording its operation so as to ensure the safety both of the material being processed and of the process operators. In this way a proper record of the operations of support facilities can be kept and, where necessary, entered into the process record of the product batches.

The equipment should be such as to ensure that both the fractionation process and the proteins are protected if the support services are interrupted. To this end, adequate spare equipment and emergency reserve systems should be available, serviced by engineering staff skilled in the maintenance and repair of such equipment.

12. Personnel

The plasma fractionation plant shall be under the direction of a designated qualified person who shall be responsible for ensuring that all operations are carried out properly and competently. The director shall have a good working knowledge of the scientific principles involved and shall be responsible for ensuring that employees are adequately trained, have adequate practical experience and are aware that accepted good practices should be applied in their work.

The personnel involved in quality-control functions shall be separate from those involved in production. The head of the quality-control department shall be responsible only to the director.

Where appropriate, personnel shall wear gowns, masks, boots, gloves and eye protectors.

Personnel should be medically examined at regular intervals. Those known to be carriers of specific pathogenic organisms that may adversely affect the product shall be excluded from the production area.

Vaccination against hepatitis B is strongly recommended for employees routinely exposed to blood or blood products.

13. Production control

13.1 *Fractionation of source materials*

The general conditions for the large-scale fractionation of source materials to prepare prophylactic or therapeutic blood products shall comply with Good Manufacturing Practices for Pharmaceutical (*7*) and Biological (*8*) Products and shall be approved by the national control authority.

Most physical and chemical techniques of protein separation may be used for the preparation of plasma fractions, provided that they yield protein preparations that have previously been shown to be safe and effective.

The fractionation procedures used shall give a good yield of products meeting the quality requirements of international or national authorities. Fractionation shall be carried out in such a manner that the risk of microbiological contamination and protein denaturation is minimized.

> The safety of fractionation steps may be increased by using protected or closed systems. Reproducibility may be increased by the use of automation.
>
> The biological characteristics of the products (such as antibody activity, biological half-life and *in vivo* recovery of the proteins) should not be affected by the fractionation procedures to the extent that they are unacceptable for clinical use.

Methods shall be used that exclude or inactivate pathogenic organisms, in particular hepatitis viruses and human retroviruses, from the final products intended for clinical use. Manufacturers shall validate the ability of their manufacturing processes to inactivate and/or remove potential contaminating viruses by the use of relevant model viruses.

There is increasing evidence that certain manufacturing procedures, coupled with strict control to ensure that the final product complies with precise specifications, result in a product free from HIV, hepatitis B and hepatitis C infectivity.

For coagulation products, viral inactivation and removal methods such as chromatography or treatment with dry heat, wet heat, steam under pressure, heated organic solvents or solvents/detergents shall be used, in combination with other methods that have been shown to be successful in reducing or eliminating the risk of HIV and hepatitis virus transmission.

Donor screening and viral inactivation procedures used in manufacturing plasma coagulation concentrates have significantly improved the safety of these products.

Fibrinogen prepared from plasma pools continues to carry a risk of infection unless it is treated to remove or inactivate viruses. Where large-pool, virally inactivated fibrinogen concentrates are not available, cryoprecipitated factor VIII prepared from individual units or small pools of plasma is preferred as a source of fibrinogen. Approximately 150 mg of fibrinogen is contained in the cryoprecipitate from one unit of plasma (200 ml) frozen within 8 h of collection from the donor.

The operating manual for the fractionation procedure shall specify the times of sampling of the products and the volumes to be taken at each stage of the process as well as the tests to be made on the samples.

Where appropriate, all materials used for fractionation shall be tested for microbiological contamination, identity, purity, endotoxin content and toxicity in accordance with *The international pharmacopoeia* (*14, 15*) or national pharmacopoeia.

Certain procedures, equipment and materials may introduce contaminants into the final product that can induce allergenic or immunogenic responses in recipients. The quantities of such contaminants in the final product shall be minimized. For example, where monoclonal antibodies are used for product purification, the residual concentration in the final product must be below clinically reactive levels.

It is advisable to use air filtration under positive pressure during fractionation, to exclude airborne allergenic dust.

13.1.1 *Preservatives and stabilizers*

No preservatives shall be added to albumin, plasma protein fraction, intravenous immunoglobulin or coagulation-factor concentrates either during fractionation or at the stage of the final bulk solution. Antibiotics shall not be used as preservatives or for any other purpose in the fractionation of plasma.

To prevent protein denaturation, stabilizers may be added. Such substances shall have been shown to the satisfaction of the national control authority not to have any deleterious effect on the final product in the amounts present and to cause no untoward reactions in humans.

Stable solutions of immunoglobulins may be prepared in approximately 0.3 mol/l glycine or 0.15 mol/l sodium chloride. In some countries, thiomersal and sodium timerfonate are not permitted as preservatives in intramuscular immunoglobulins.

13.2 ***Storage and control of source materials***

At all stages of the manufacturing process, the source materials and resulting fractions shall be stored at temperatures and under conditions

shown to prevent further contamination and the growth of micro-organisms, to protect the identity and the integrity of the proteins and to preserve the biological activity and safety of the products.

If similar materials are stored together, the places allocated to them shall be clearly demarcated.

All source materials and resulting fractions shall be fully identified at all times; such identification shall include the batch number of all in-process fractions and final containers awaiting labelling.

13.2.1 *In-process control*

Source materials are subject to biological variability and the products resulting from protein separation will contain various amounts of other protein components of plasma. It is essential, therefore, to establish a monitoring system such that the safe operating limits of each process are maintained.

The main information collected is on variations in physical conditions (temperature, pH, ionic strength, timing, etc.) and in the number and species of contaminating organisms.

Owing to the numerous and interdependent factors involved, there are no universally accepted specifications for such in-process quality-assurance systems. For this reason, the information collected should be combined with data from previous experience with the same manufacturing process to ensure production control appropriate to the quality requirements of the final product.

13.2.2 *Record-keeping*

Records shall be kept of the performance of all steps in the manufacture, quality control and distribution of large-pool blood products and related substances (*7, 8*).

These records shall:

- be original (not a transcription), indelible, legible and dated;
- be made at the time that the specific operations and tests are performed;
- identify the person recording the data as well as the person checking them or authorizing the continuation of processing;
- be detailed enough to allow all the relevant procedures performed to be clearly reconstructed and understood;
- permit the tracing of all successive steps and identify the relationships between dependent procedures, products and waste materials;
- be maintained in an orderly fashion that will permit the retrieval of data for a period consistent with shelf-lives and the legal requirements of the national control authority and, if necessary, allow a prompt and complete recall of any particular lot;
- show the lot numbers of the materials used for specified lots of products;
- indicate that processing and testing were carried out in accordance with procedures established and approved by the designated responsible authority.

14. Control of albumin and plasma protein fraction

Source materials should be processed in such a manner that the albumin in the solutions manufactured will be changed as little as possible and will not cause undesirable reactions in the recipients. Source materials may contain either vasoactive substances or substances capable of generating or releasing endogenous vasoactive substances. Such substances may also be formed in the course of fractionation, and consequently contaminate the albumin and plasma protein fraction. To guard against this possibility, adequate in-process controls and the testing before release for prekallikrein activator activity are mandatory for albumin solutions of purity less than 95% (such as plasma protein fraction) containing 35–50 g of protein per litre. Such testing is also recommended for highly purified albumin products (purity greater than 95%).

Within 24 h of the start of filling, albumin and plasma protein fraction in solution shall be heated in the final container to 60 ± 0.5 °C and maintained at that temperature for not less than 10 h but not more than 11 h by a method that ensures uniform heat distribution throughout the batch. Although pasteurization at the final bulk stage may be possible, this approach requires careful validation before use.

Special attention should be given to microbial contamination of source material and intermediates, since soluble microbial substances, especially endotoxins, may accumulate in the finished albumin solution. In addition, it is possible that small amounts of endotoxin, present even in products for which satisfactory results have been obtained in tests for pyrogens, may have a cumulative effect in recipients receiving large product volumes in relatively short periods of time, as, for example, in therapeutic plasma exchange.

In some countries, information is being collected about the usefulness of quantitative *Limulus* assays for the presence of endotoxin.

The in-process controls should be capable of detecting contamination with bacteria and moulds. In addition, care should be taken to ensure, by a method that shall be validated, that all equipment and reagents used in the manufacturing process are scrupulously clean and free from toxic materials.

14.1 *Stability of albumin solutions*

The stability of solutions of albumin and plasma protein fraction (that have been heated for 10–11 h at 60 °C) shall be tested by heating adequate samples at 57 °C for 50 h. The test solutions shall remain visually unchanged when compared to control samples that have been heated for only 10–11 h at 60 °C.

The thermal stability of albumin solutions shall be taken into consideration by the national control authority in determining the expiry dates.

The physicochemical quality of stored albumin solutions, as measured by the formation of dimers and particularly polymers, is influenced by:

- the quality of the starting plasma;
- the quality of the fractionation, particularly with respect to the degree of purity achieved and the number of reprecipitation and reheating procedures involved; and
- the storage conditions with respect not only to temperature and time but also to the physical state and concentration of the solutions.

With regard to the thermal stability of albumin solutions, the following general statements may be made:

- The addition of stabilizing chemicals is necessary. Commonly used products are sodium octanoate and sodium acetyltryptophanate.
- Albumin prepared from aged liquid or dried plasma is less stable than albumin made from fresh-frozen plasma.
- Reprocessing steps, such as reprecipitation and reheating, may reduce the stability of albumin solutions.
- On long-term storage, albumin solutions are more stable at 5 ± 3 °C than at 32–35 °C. Long-term storage above 30 °C should be avoided.

14.2 *Control of bulk material*

14.2.1 *Tests on bulk material*

Tests on the bulk powder or solution shall be made if the manufacturer sends the material to another institution for further processing. Samples for these tests shall be taken under conditions that do not impair the quality of the bulk material. Tests shall be carried out on a specially dissolved sample processed to a stage equivalent to the final product, after sterilization by filtration. The tests shall be those listed in sections 14.3.2 to 14.3.7 inclusive.

14.2.2 *Storage*

The bulk material shall be stored as liquid or powder in sealed containers under conditions that minimize denaturation and the multiplication of microbial agents.

14.3 *Control of the final bulk solution*

14.3.1 *Preparation*

The final bulk solution shall be prepared from bulk powder or by the dilution of concentrates by a method approved by the national control authority. It shall meet all of the requirements of sections 14.3.2 to 14.3.7 inclusive.

14.3.2 *Concentration and purity*

The albumin concentration in final bulk albumin solutions shall be between 35 and 265 g/l. Not less than 95% of the proteins present shall be albumin, as determined by a suitable electrophoretic method after the sample has been heated for 10–11 h at 60 °C.

The protein concentration in final bulk solutions of plasma protein fraction shall be at least 35 g/l. Plasma protein fraction shall contain at least 83% albumin and not more than 17% globulins. Not more than 1% of the protein in plasma protein fraction shall be γ-globulin.

14.3.3 *Hydrogen ion concentration*

The final bulk solution, diluted with 0.15 mol/l sodium chloride to give a protein concentration of 10 g/l, shall, when measured at a temperature of 20–27 °C, have a pH of 6.9 ± 0.5 (albumin) or 7.0 ± 0.3 (plasma protein fraction).

In some countries, different ranges of pH values and temperatures are permitted.

14.3.4 *Sterility and safety*

The final bulk shall be sterile. If required by the national control authority, it shall be tested for sterility; samples shall be taken for such testing in a manner that does not compromise the sterility of the bulk material. Part A, section 5, of the revised Requirements for Biological Substances No. 6 (General Requirements for the Sterility of Biological Substances) (*9*, p. 48) shall apply.

14.3.5 *Sodium content*

The final bulk solutions of albumin and plasma protein fraction shall have a maximum sodium concentration of 160 mmol/l.

14.3.6 *Potassium content*

The final bulk solutions of albumin and plasma protein fraction shall have a maximum potassium concentration of 2.0 mmol/l.

14.3.7 *Aluminium content*

The final bulk solutions of albumin and plasma protein fraction shall have a maximum aluminium concentration of 7.5 μmol/l (200 μg/l).

14.4 ***Filling and containers***

The requiremens concerning filling and containers given in Good Manufacturing Practices for Biological Products (*8*) shall apply.

Special attention shall be paid to the requirement that solutions of albumin and plasma protein fraction in the closed final containers shall be heated to inactivate any infectious agents that may be present (see section 14, paragraph 2). In order to prevent protein denaturation, a stabilizer shall be added to albumin solution before heating (see section 13.1.1).

In some countries, the national control authority may authorize an interval longer than 24 h between filling and heating to 60 °C.

14.5 *Control tests on the final product*

The tests specified below shall be performed on representative samples from every filling lot. If the product is processed further after filling, e.g. by freeze-drying, the tests shall be performed on samples from each drying chamber.

14.5.1 *Identity test*

An identity test shall be performed on at least one labelled container from each filling lot to verify that the preparation is of human origin. The test shall be one approved by the national control authority. Additional tests shall be made to determine that the protein is predominantly albumin or plasma protein fraction as appropriate. The tests mentioned in section 14.3.2 shall be used.

14.5.2 *Protein concentration and purity*

The protein concentration and purity of each filling lot shall be within the limits prescribed in section 14.3.2.

Tests to determine the concentration of additives (such as polyethylene glycol, porcine enzymes and reducing and alkylating agents) used during production shall be carried out if required by the national control authority.

14.5.3 *Sterility test*

Each filling lot shall be tested for sterility. Part A, section 5, of the revised Requirements for Biological Substances No. 6 (General Requirements for the Sterility of Biological Substances) (*9*, p. 48) shall apply. Samples for sterility testing shall be taken from final containers selected at random after heating at 60 °C for 10–11 h.

In one country, the sterility test is carried out at least 10 days after heating at 60 °C for 10 h. In some countries, the sterility test is carried out both before and after heating at 60 °C for 10 h.

14.5.4 *General safety test*

In some countries a general safety test may be required, whereby each filling lot is tested for extraneous toxic contaminants by appropriate tests involving injection into mice and guinea-pigs. The injection shall cause neither significant untoward reactions nor death within an observation period of seven days. The tests shall be approved by the national control authority.

The tests generally used are the intraperitoneal injection of 0.5 ml into each of at least two mice weighing approximately 20 g and the injection of 5.0 ml into each of at least two guinea-pigs weighing approximately 350 g. In some countries, if one of the animals dies or shows signs of ill-health, such as weight loss, during a specified period, the test is repeated. The substance passes the test if none of the animals of the second group dies or shows signs of ill-health, such as weight loss, during that period.

14.5.5 *Freedom from pyrogenicity*

Each filling lot shall be tested for pyrogenicity by the intravenous injection of the test dose into three or more rabbits that have not previously received blood products. In general, the dose shall be at least equivalent proportionally, on a rabbit body-weight basis, to the maximum single human dose recommended, but not more than 10 ml/kg of body weight. For albumin at concentrations of 200 g/l and 250 g/l, the test dose for each rabbit shall be at least 3 ml/kg of body weight, and for albumin at concentrations of 35 g/l and 50 g/l and plasma protein fraction, 10 ml/kg of body weight.

A filling lot shall pass the test if it satisfies the requirements specified by the national control authority.

14.5.6 *Moisture content*

The residual moisture content shall, where appropriate, be determined by a method approved by the national control authority.

The methods in use are: (a) drying over phosphorus pentoxide for at least 24 h at a pressure not exceeding 2.7 Pa (0.02 mmHg); and (b) the Karl Fischer method.

The acceptable moisture content shall be determined by the national control authority.

14.5.7 *Prekallikrein activator*

An assay shall be performed for prekallikrein activator. The product shall contain not more than 35 IU of prekallikrein activator per ml.

14.5.8 *Hydrogen ion concentration*

The final product, reconstituted if necessary and diluted with 0.15 mol/l sodium chloride to give a protein concentration of 10 g/l, shall, when measured at a temperature of 20–27 °C, have a pH of 6.9 ± 0.5 (albumin) or 7.0 ± 0.3 (plasma protein fraction).

In some countries, different ranges of pH values are permitted.

14.5.9 *Absorbance*

A sample taken from the final solutions of albumin and plasma protein fraction, when diluted with water to a concentration of 10 g/l of protein and placed in a cell with a 1-cm light path, shall have an absorbance not exceeding 0.25 when measured in a spectrophotometer set at 403 nm.

14.5.10 *Inspection of filled containers*

All final containers shall be inspected for abnormalities, such as non-uniform colour, turbidity, microbial contamination and the presence of atypical particles, after storage at 20–35 °C for at least 14 days following heat treatment at 60 °C for 10 h. Containers showing abnormalities shall not be distributed.

The normal colour of albumin solutions may range from colourless to yellow or green to brown.

When turbidity or non-uniform colour raises the possibility of microbial contamination, testing should be done to isolate and identify the microorganisms.

14.6 ***Records***

The requirements of Good Manufacturing Practices for Biological Products (*8*, pages 27–28) shall apply.

14.7 ***Samples***

The requirements of Good Manufacturing Practices for Biological Products (*8*, page 29, paragraph 9.5) shall apply.

14.8 ***Labelling***

The requirements of Good Manufacturing Practices for Biological Products (*8*, pages 26–27) and the national control authority's requirements for parenteral solutions shall apply.

In addition, the label on the container should state:

– the type of source material,
– the protein concentration,
– the oncotic equivalent in terms of plasma,
– that preservatives are absent
– the warning "Do not use if turbid",
– the sodium and potassium concentrations.

14.9 ***Distribution and shipping***

The requirements of Good Manufacturing Practices for Biological Products (*8*) shall apply.

14.10 ***Storage and shelf-life***

The requirements of Good Manufacturing Practices for Biological Products (*8*, pages 26–27) shall apply.

Final containers of albumin solution shall have a maximum shelf-life of three years if they are stored at or below 30 °C, and of five years if they are stored at 5 ± 3 °C.

Other storage conditions and shelf-lives may be approved by the national control authority.

Final containers of plasma protein fraction solution shall have a maximum shelf-life of three years if they are stored at or below 30 °C, and of five years if they are stored at 5 ± 3 °C.

Other storage conditions and shelf-lives may be approved by the national control authority.

15. Control of immunoglobulins

The final bulk solution of normal immunoglobulin shall be made from material from at least 1000 donors. If normal immunoglobulin is to be used for preventing or treating a particular infection, the titre of specific antibody should be measured.

For normal immunoglobulins, a large number of donors are needed if the final product is to contain adequate amounts of the various desired antibodies.

For specific immunoglobulins, whether intended for intravenous or intramuscular injection, the number of donors represented is less important because the requirement for specific antibody in the final product will be defined.

The immunoglobulin concentration in the final bulk of normal and specific immunoglobulin preparations for intramuscular use shall be 100–180 g/l. Concentrations lower than 100 g/l shall require the approval of the national control authority.

The immunoglobulin concentration in the final bulk of intravenous immunoglobulin shall be at least 30 g/l. If, in a specific immunoglobulin preparation, the concentration is lower than 30 g/l, it shall require the approval of the national control authority.

The immunoglobulin preparation shall be composed of not less than 90% of immunoglobulin, as determined by a method approved by the national control authority.

Tests shall be conducted on each filling lot of immunoglobulin solution to determine the proportion of aggregated and fragmented immunoglobulin. The recommended distribution shall be that at least 90% of the protein, other than proteins added as stabilizers to intravenous immunoglobulins, shall have the molecular size of immunoglobulin monomer and dimer. Not more than 10% shall consist of split products together with aggregates (oligomers of relative molecular mass equal to or greater than that of immunoglobulin trimer). This requirement shall not apply to products deliberately fragmented. The tests and limits shall be approved by the national control authority. Of the material having the molecular size of immunoglobulin monomer and dimer, most will consist of monomer. If a minimum level of monomer *per se* is to be established, the time and temperature at which samples must be incubated before analysis shall be specified.

Gel-permeation chromatography and high-performance exclusion chromatography are useful techniques for determining molecular size distribution and can be standardized for making these measurements.

For intravenous immunoglobulin, the following tests shall be performed on a sample from each filling lot:

- A test for hypotensive activity.

An appropriate test is that for prekallikrein activator content. In some countries the kallikrein test is also used.

- **A test for anticomplement activity.**

  Several methods are available. The test method used and the maximum level of anticomplement activity permitted should be approved by the national control authority.

- **A test for haemagglutinins by the antiglobulin (Coombs) technique.**

  In such tests, group O D(Rh_o)-positive cells should be used to test for anti-D (anti-Rh_o); group A and group B D(Rh_o)-negative cells should be used for anti-A and anti-B, respectively.

  The purpose of the test is to ensure that the use of the product will not give rise to haemolytic reactions. The upper limit of activity should be specified by the national control authority.

15.1 *Potency of normal immunoglobulins*

A 160 g/l solution of normal immunoglobulin shall be prepared from final bulk solution by a method that has been shown to be capable of concentrating, by a factor of 10 from source material, at least two different antibodies, one viral and one bacterial, for which an international standard or reference preparation is available (*16*) (e.g. antibodies against poliomyelitis virus, measles virus, streptolysin O, diphtheria toxin, tetanus toxin, staphylococcal α-toxin).

For immunoglobulins formulated at an immunoglobulin concentration lower than 16%, the concentrating factor for antibodies from source material may be proportionally lower.

The immunoglobulin solution shall be tested for potency at the concentration at which it will be present in the final container.

Since preparations of normal immunoglobulins produced in different countries can be expected to differ in their content of various antibodies, depending upon the antigenic stimulation to which the general population has been subjected (either by natural infection or by deliberate immunization), at least two antibodies should be chosen for the potency test by the national control authority. The final product passes the test if it contains at least the minimum antibody levels required by the national control authority.

15.2 *Potency of specific immunoglobulins*

The potency of each final lot of specific immunoglobulin shall be tested with respect to the particular antibody that the preparation has been specified to contain. For intramuscular immunoglobulins, the following levels shall apply:

- For tetanus immunoglobulin, at least 100 IU/ml of tetanus antitoxin, as determined by a neutralization protection test in animals or by a method shown to be equivalent.
- For rabies immunoglobulin, at least 100 IU/ml of anti-rabies antibody,

as determined by an appropriate neutralization test in animals or by a method shown to be equivalent.

- For hepatitis B immunoglobulin, at least 100 IU/ml of anti-hepatitis antibody.
- For varicella zoster immunoglobulin, at least 100 IU/ml of anti-varicella zoster antibody, as measured by a comparative enzyme-linked immunosorbent assay or by a method shown to be equivalent.
- For anti-D (anti-Rh_0) immunoglobulin, the estimated potency shall be expressed in International Units and shall be not less than 90% and not more than 120% of the stated potency, and the fiducial limits of error shall be within 80% and 125% of the estimated potency.

The national control authority shall specify the antibody limits for other immunoglobulins.

After the potency tests, a test for immunoglobulin subclass may be performed. Different manufacturing steps have been shown to reduce the concentration of specific immunoglobulin subclasses (e.g. IgG1, IgG2, IgG3 and IgG4) in immunoglobulin preparations. The distribution of the four subclasses of IgG may be a factor in the efficacy of intravenous immunoglobulin preparations, since specific antibodies belonging to particular subclasses have been identified as being important in several infectious diseases.

In some countries the distribution of IgG subclasses has been measured by radial immunodiffusion. Enzyme-linked immunosorbent assays have also been described, and may be used if properly validated. Assays should be calibrated against the appropriate international reference materials.

15.3 *Sterility and safety*

Each filling lot shall be tested for sterility. Part A, section 5, of the revised Requirements for Biological Substances No. 6 (General Requirements for the Sterility of Biological Substances) (*9*, p. 48) shall apply.

In some countries a general safety test may be required, whereby each filling lot is tested for extraneous toxic contaminants by appropriate tests involving injection into mice and guinea-pigs. The injection shall cause neither significant toxic reactions nor death within an observation period of seven days. The tests shall be approved by the national control authority.

The tests generally used are the intraperitoneal injection of 0.5 ml into each of at least two mice weighing approximately 20 g and the injection of 5.0 ml into each of at least two guinea-pigs weighing approximately 350 g. In some countries, if one of the animals dies or shows signs of ill-health, such as weight loss, during a specified period, the test is repeated. The substance passes the test if none of the animals of the second group dies or shows signs of ill-health, such as weight loss, during that period.

15.4 *Identity test*

An identity test shall be performed on at least one labelled container from each filling lot to verify that the preparation is of human origin. The test shall be one approved by the national control authority.

Additional tests shall be made to determine that the protein is predominantly immunoglobulin.

The methods in most common use are radial immunodiffusion and electrophoresis.

15.5 *Freedom from pyrogenicity*

Each filling lot shall be tested for pyrogenicity by the intravenous injection of the test dose into three or more rabbits that have not previously received blood products. In general, the dose shall be at least equivalent proportionally, on a rabbit body-weight basis, to the maximum single human dose recommended, but not more than 10 ml/kg of body weight. The recommended test doses are 1 ml/kg and 10 ml/kg of body weight for intramuscular and intravenous preparations, respectively.

A filling lot shall pass the test if it satisfies the requirements specified by the national control authority.

15.6 *Moisture content*

The residual moisture content of a sample from each filling lot shall, where appropriate, be determined by a method approved by the national control authority.

The methods in use are: (a) drying over phosphorus pentoxide for at least 24 h at a pressure not exceeding 2.7 Pa (0.02 mmHg); and (b) the Karl Fischer method.

The acceptable moisture content shall be determined by the national control authority.

15.7 *Hydrogen ion concentration*

The final product, reconstituted if necessary and diluted with 0.15 mol/l sodium chloride to give a protein concentration of 10 g/l, should, when measured at a temperature of 20–27 °C, have a pH of 6.9 ± 0.5.

In some countries, a different range of pH values is permitted for intravenous immunoglobulins.

15.8 *Stability*

For immunoglobulin solutions, a stability test shall be performed on each filling lot by heating an adequate sample at 37 °C for four weeks. No gelation or flocculation shall occur.

Alternatively (or in addition), the molecular size distribution of the immunoglobulin or assays of enzymes such as plasmin (fibrinolysin) may be used, when shown to predict stability reliably and when approved by the national control authority.

15.9 *Records*

The requirements of Good Manufacturing Practices for Biological Products (*8*, pages 27–28) shall apply.

15.10 *Samples*

The requirements of Good Manufacturing Practices for Biological Products (*8*, page 29, paragraph 9.5) shall apply.

15.11 *Labelling*

The requirements of Good Manufacturing Practices for Biological Products (*8*, pages 26–27) shall apply.

In addition, the label on the container shall state:

- the type of source material;
- the protein concentration;
- the concentration of preservative, if any;
- "For intramuscular use only" (if the immunoglobulins are not specially prepared for intravenous use);
- "For intravenous use", when appropriate;
- for specific immunoglobulin, the content of specific antibody expressed in International Units or equivalent national units;
- for freeze-dried preparations, the name and volume of reconstituting liquid to be added.

The label on the package or the package insert shall show:

- the approximate concentration of electrolytes and excipients and, for intravenous preparations, the approximate osmolality;
- the buffering capacity when the pH of the diluted product is lower than that specified in section 15.7;
- the concentration of preservative, if any;
- the recommended dose for each particular disease or condition;
- the warning "Do not use if turbid";
- the sodium and potassium concentrations (if the immunoglobulin is intended for intravenous use).

15.12 *Distribution and shipping*

The requirements of Good Manufacturing Practices for Biological Products (*8*) shall apply.

15.13 *Storage and shelf-life*

The requirements of Good Manufacturing Practices for Biological Products (*8*, pages 26–27) shall apply.

Liquid immunoglobulin shall be stored at 5 ± 3 °C and shall have a shelf-life of not more than three years. Freeze-dried preparations shall be stored below 25 °C and shall have a shelf-life of not more than five years.

Other storage conditions and shelf-lives may be approved by the national control authority.

16. Control of preparations of coagulation-factor concentrates (factor VIII, factor IX and fibrinogen)

Factor VIII preparations are available as both frozen products and freeze-dried concentrates. The frozen products are usually derived from a single donation and consist of the cryoprecipitated factor VIII from the donor concerned prepared in a closed separation system. The control of this product and the freeze-dried product from fewer than 10 plasma donations is covered in Part B, section 7.8.1.

Generally, the small-pool product undergoes little or no purification and is handled and subdivided in such a way that many control tests are inappropriate. However, freeze-dried factor VIII concentrates prepared from more than 10 donations may be purified.

Source material for factor VIII preparations shall meet the general criteria for donor selection and testing for disease markers as specified in Parts A and B. It shall preferably be plasma frozen within 8 h of collection or frozen cryoprecipitate. Such material shall be kept frozen at such a temperature that the activity of the factor VIII is maintained.

16.1 *Tests on final containers*

16.1.1 *Sterility and safety*

Each filling lot shall be tested for sterility. Part A, section 5, of the revised Requirements for Biological Substances No. 6 (General Requirements for the Sterility of Biological Substances) (*9*, p. 48) shall apply.

In some countries a general safety test may be required, whereby each filling lot is tested for extraneous toxic contaminants by appropriate tests involving injection into mice and guinea-pigs. The injection shall cause neither significant toxic reactions nor death within an observation period of seven days. The tests shall be approved by the national control authority.

The tests generally used are the intraperitoneal injection of 0.5 ml into each of at least two mice weighing approximately 20 g and the injection of 5.0 ml into each of at least two guinea-pigs weighing approximately 350 g. In some countries, if one of the animals dies or shows signs of ill-health, such as weight loss, during a specified period, the test is repeated. The substance passes the test if none of the animals of the second group dies or shows signs of ill-health, such as weight loss, during that period. For factor VIII and factor IX concentrates, the test dose should not exceed 500 IU of the coagulation factor per kg of body weight of the test animal.

16.1.2 *Freedom from pyrogenicity*

Each filling lot shall be tested for pyrogenicity by the intravenous injection of the test dose into three or more rabbits that have not previously received blood products. In general, the dose shall be at least equivalent

proportionally, on a rabbit body-weight basis, to the maximum single human dose recommended, but not more than 10 ml/kg of body weight.

The following test doses are suggested: factor VIII, 10 IU/kg of body weight; factor IX, 50 IU/kg of body weight; and fibrinogen, 30 mg/kg of body weight.

16.1.3 *Solubility and clarity*

Factor VIII preparations shall dissolve in the solvent recommended by the manufacturer within 30 min when held at a temperature not exceeding 37 °C. Factor IX preparations shall dissolve in the solvent recommended by the manufacturer within 15 min when held at 20–25 °C. The solutions, when kept at room temperature, shall not show any sign of precipitation or gel formation within 3 h of dissolution of the coagulation factors.

16.1.4 *Protein content*

The amount of protein in a final container shall be determined by a method approved by the national control authority.

16.1.5 *Additives*

Tests to determine the concentration of additives (such as heparin, polyethylene glycol, sodium citrate and glycine) used during production shall be carried out if required by the national control authority.

16.1.6 *Moisture content*

The residual moisture content shall be determined by a method approved by the national control authority. The acceptable moisture content shall be determined by the national control authority.

The methods available are: (a) drying over phosphorus pentoxide for 24 h at a pressure not exceeding 2.7 Pa (0.02 mmHg); and (b) the Karl Fischer method.

16.1.7 *Hydrogen ion concentration*

When the product is dissolved in a volume of water equal to the volume stated on the label, the pH of the resulting solution shall be 7.2 ± 0.4.

In some countries, different pH values are approved.

16.2 Test applicable to factor VIII concentrates

Each filling lot shall be assayed for factor VIII activity by a test approved by the national control authority, using a standard calibrated against the International Standard for Blood Coagulation Factor VIII: Concentrate.

The national standard and the manufacturer's house standard should be a concentrate rather than a plasma because the former has better long-term stability and provides more homogeneous assay results.

The specific activity shall be at least 500 IU/g of protein. The estimated potency shall be not less than 80% and not more than 125% of the stated potency. The confidence limits of error shall be not less than 64% and not more than 156% of the estimated potency.

16.3 *Tests applicable to factor IX concentrates*

16.3.1 *Potency*

Each filling lot shall be assayed for factor IX activity by a test approved by the national control authority, using a standard calibrated against the International Standard for Human Blood Coagulation Factors II, IX, and X in Concentrates.

Other coagulation factors may also be present in the final product, depending on the method of production, and products shall be assayed for all coagulation factors claimed to be present at a therapeutic level, including factors II, VII and X. The assay methods used for these factors shall be approved by the national control authority.

16.3.2 *Presence of activated coagulation factors*

A test for the presence of activated coagulation factors shall be carried out by a method approved by the national control authority.

In some countries, the non-activated partial thromboplastin times of normal plasma are measured after the addition of an equal volume of a number of different dilutions of the product under test.

In some countries, a test for the presence of thrombin is carried out by mixing equal volumes of the product under test and fibrinogen solution. The mixture is held at 37 °C and should not coagulate within 6 h. The usual range of concentrations of fibrinogen solution is 3–10 g/l.

16.3.3 *Alloantibodies*

A test shall be made for the presence of alloantibodies A and B by a method approved by the national control authority.

It is not possible to be specific about the tests for alloantibodies or to specify an upper limit for the titre.

16.4 *Test applicable to fibrinogen*

Each filling lot shall be assayed for clottable protein by a test approved by the national control authority.

Not less than 70% of the total protein should be clottable by thrombin.

16.5 *Identity test*

An identity test shall be performed on at least one labelled container from each filling lot of coagulation-factor concentrate to verify that the preparation is of human origin. The test shall be one approved by the national control authority.

For albumin and plasma protein fraction, additional tests shall be made to determine that the protein is predominantly albumin.

The methods in most common use are radial immunodiffusion and electrophoresis.

16.6 ***Records***

The requirements of Good Manufacturing Practices for Biological Products (*8*, pages 27–28) shall apply.

16.7 ***Samples***

The requirements of Good Manufacturing Practices for Biological Products (*8*, page 29, paragraph 9.5) shall apply.

16.8 ***Labelling***

The requirements of Good Manufacturing Practices for Biological Products (*8*, pages 26–27) shall apply.

In addition, the label on the container shall state:

- the content of the coagulation factor expressed in International Units, where they exist;
- the amount of protein in the container;
- the volume of diluent needed for reconstitution;
- a reference to a package insert giving instructions for use, warnings about the possible transmission of infectious agents and precautions.

16.9 ***Distribution and shipping***

The requirements of Good Manufacturing Practices for Biological Products (*8*) shall apply.

16.10 ***Storage and shelf-life***

The requirements of Good Manufacturing Practices for Biological Products (*8*, pages 26–27) shall apply.

Final containers of freeze-dried preparations of factor VIII and factor IX shall have a maximum shelf-life of two years if they are stored at 5 ± 3 °C. Final containers of fibrinogen shall have a maximum shelf-life of five years if they are stored at 5 ± 3 °C.

Other storage conditions and shelf-lives may be approved by the national control authority provided that they are consistent with the data on the stability of the products.

Part D. National control requirements

17. General

The general requirements for control laboratories in the Guidelines for National Authorities on Quality Assurance for Biological Products (*6*) shall apply.

The national control authority shall provide the standards and reference preparations necessary for the quality control of human blood and blood

products. Where appropriate, these standards should be calibrated against the relevant International Standard.

The national control authority shall have authority to approve the production and control methods used and settle all matters left for its decision or approval in Parts A, B and C.

The national control authority shall also have authority to approve the use of materials that carry potential risk and shall approve any new method of production and the preparation of any new product.

New products or products prepared by new production methods may be monitored to confirm their efficacy and safety.

18. Release and certification

Human blood and blood products shall be released only if they satisfy the requirements of Parts A, B and C, wherever applicable.

A certificate signed by the appropriate official of the national control authority shall be provided at the request of the manufacturing establishment and shall state whether the product in question meets all national requirements as well as Parts A, B and C (whichever is relevant) of the present Requirements. The certificate shall also state the date of the last satisfactory potency test performed by the manufacturer, if applicable, the number under which the lot is released, and the number appearing on the labels of the containers. In addition, a copy of the official national release document shall be attached.

The purpose of this certificate is to facilitate the exchange of human blood and blood products between countries.

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Appendix
Summary protocol for collection of source material

1. Name and address of collecting centre ______________________

____

2. Source material ______________________

3. Details of single donations, where applicable:
   (a) Donor identification ______________________
   (b) Date of collection ______________________
   (c) Volume in container ______________________
   (d) Results of tests for HBsAg ______________________
   (e) Results of tests for anti-HIV ______________________
   (f) Results of tests for anti-HCV ______________________
   (g) If applicable, results of tests for antibody to hepatitis B core antigen ______________________
   (h) If applicable, results of tests for alanine aminotransferase ______________________

4. Special information:
   (a) Anticoagulant used ______________________
   (b) Was the material collected for special purposes (e.g. as a source of specific antibodies)? ______________________
   (c) Precautions to be taken when using the material ______________________

5. Conditions of storage ______________________

6. Does the donation comply with existing agreements between the supplier and manufacturer? ______________________

7. Does the donation comply with the Requirements for the Collection, Processing and Quality Control of Blood, Blood Components and Plasma Derivatives published by WHO? ______________________

Name and signature of responsible person ______________________

Date ______________________

8

STANDARD OPERATING PROCEDURE

____

(Name of the Blood Centre)

Number	Effective Date	Pages	Author	Authorised by
SP 001		3		
Version	**Review Period**	**No. of Copies**	**Approved by**	**Date**
1	1 Year			

LOCATION	SUBJECT
Donor Room	Criteria for Donor Selection
FUNCTION	**DISTRIBUTION**
Assessing suitability of donor for blood donation	- Medical Officer in charge of Donor Area - Master File

1. SCOPE & APPLICATION

This SOP describes the criteria for a donor to be accepted for blood donation, for ensuring safety of donor as well as recipient. The purpose of donor selection is to identify any factors that might make an individual unsuitable as a donor, either temporarily or permanently.

2. RESPONSIBILITY

The Medical Officer is responsible for determining the suitability of donor for blood donation. He/She should confirm that the criteria are fulfilled after evaluation of health history questionnaire and medical examination including the results of pre donation screening tests.

3. REFERENCES

Technical Manual of American Association of Blood Banks- 13th edition, 1999 pgs 90-97, 103-110.

4. MATERIAL REQUIRED

- Donor Questionnaire
- Donor Card

5. PROCEDURE

CRITERIA FOR SELECTION OF BLOOD DONORS

A. Accept only voluntary/replacement non-remunerated blood donors if following criteria are fulfilled.

The interval between blood donations should be no less than three months. The donor shall be in good health, mentally alert and physically fit and shall not be a jail inmate or a person having multiple sex partners or a drug-addict. The donors shall fulfill the following requirements, namely:-

1. The donor shall be in the age group of 18 to 60 years
2. The donor shall not be less than 45 kilograms
3. Temperature and pulse of the donor shall be normal
4. The systolic and diastolic blood pressures are within normal limits without medication
5. Haemoglobin shall not be less than 12.5 g/dL
6. The donor shall be free from acute respiratory diseases
7. The donor shall be free from any skin disease at the site of phlebotomy
8. The donor shall be free from any disease transmissible by blood transfusion, in so far as can be determined by history and examination indicated above
9. The arms and forearms of the donor shall be fee from skin punctures or scars indicative of professional blood donors or addiction of self-injected narcotics

B. Defer the donor for the period mentioned as indicated in the following table:

CONDITIONS	PERIOD OF DEFERMENT
Abortion	6 months
History of blood transfusion	6 months
Surgery	12 months
Typhoid fever	12 months after recovery
History of Malaria duly treated	3 months (endemic) 3 years (non endemic area)
Tattoo	6 months
Breast feeding	12 months after delivery
Immunization (Cholera, Typhoid, Diphtheria, Tetanus, Plague, Gammaglobulin)	15 days
Rabies vaccination	1 year after vaccination
Hepatitis in family or close contact	12 months
Hepatitis Immune globulin	12 months

C. Defer the donor permanently if suffering from any of the following diseases:

1. Cancer
2. Heart disease
3. Abnormal bleeding tendencies
4. Unexplained weight loss
5. Diabetes
6. Hepatitis B infection
7. Chronic nephritis

- Signs and symptoms, suggestive of AIDS

9. It is important to ask donors if they have been engaged in any risk behaviour. Allow sufficient time for discussion in the private cubicle. Try and identify result-seeking donors and refer them to VCTC (Voluntary Counseling and Testing Center). Reassure the donor that strict confidentially is maintained.
10. Liver disease
11. Tuberculosis
12. Polycythemia Vera
13. Asthma
14. Epilepsy
15. Leprosy
16. Schizophrenia
17. Endocrine disorders

D. Private interview:

A detailed sexual history should be taken. Positive history should be recorded on confidential notebook.

E. Informed consent:

Provide information regarding:

1. Need for blood
2. Need for voluntary donation
3. Regarding transfusion transmissible infections
4. Need for questionnaire and honest answers
5. Safety of blood donation
6. How the donated blood is processed and used
7. Tests carried out on donated blood

N.B. This gives the donor an opportunity to give his/her consent if they feel they are safe donors

* Request the donors to sign on the donor card indicating that he is donating voluntarily.

6. DOCUMENTATION

Enter all details in the donor questionnaire form/card and computer