

LONG-TERM STORAGE OF IgG AND IgM ON FILTER PAPER FOR USE IN PARASITIC DISEASE SEROEPIDEMIOLOGY SURVEYS¹

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To help investigate how long IgG and IgM antibodies can be stored on filter paper, blood samples were obtained from 23 subjects, were stored on filter paper and in three other ways for 18 months, and were periodically tested by immunofluorescence and radial immunodiffusion to determine the samples' mean antibody concentrations and geometric mean antibody titers. The following article describes this investigation and reports its findings.

Introduction

Seroepidemiologic surveys are usually conducted in areas where transportation facilities and basic utilities (including electric power) tend to be inadequate, and where the climate presents a wide range of temperature and humidity fluctuations. In such circumstances, providing adequate conditions for the collection and storage of serum samples may pose a special problem for investigators.

Storage of frozen sera may be a problem if suitable containers such as deep-freezes are not available or if power failures are frequent. Moreover, long-term storage of frozen sera in test tubes causes proteins to be deposited at the bottom of the test tubes; and, upon solubilization, sample turbidity indicates that some degree of protein denaturation took place. This is a common observation in serology laboratories everywhere.

The addition of equal amounts of neutral glycerin to serum samples prevents freezing and maintains proteins in solution at temperatures as low as -20°C ; it also has a bactericidal effect that prevents bacterial contamination (1). As a result, serum samples thus diluted with glycerin may be transported long distances without alteration (2).

Freeze-drying does not protect serum samples from denaturation. Depending on storage temperature, 10S gamma globulin aggregates are formed first, followed by formation of 5S aggregates, with concomitant lowering of the 10S component's concentration as reported by James et al. (3). Such changes are due to activation of proteolytic enzymes such as plasmin that are present in the samples (4) and act on the C-terminal region of the antibody molecule.

Since the time when filter paper was first

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proposed for blood collection and storage (5, 6), some limitations have become apparent. Among other things, there is a need for precise evaluation of the serum dilution represented by the eluate; discrepancies have been found between eluate and serum titers (7, 8); and losses in antibody activity are known to occur as a result of temperature variations, humidity variations, or both (7, 9).

When most papers dealing with the problems of collecting and storing blood on filter paper were written, either immunofluorescence tests were still done with antigammaglobulin conjugates (6, 8-10), or else the techniques employed (such as hemagglutination for diagnosis of Chagas' disease) revealed only IgG antibodies (11, 12). Therefore, any differences between the IgG and IgM eluted went unnoticed.

In 1970, however, a group of WHO experts raised the question of whether IgM could be eluted from filter paper as thoroughly as IgG (13). In this same vein, Cunningham et al. (14) had reported earlier that African trypanosomiasis sera stored for 48 days at -20°C displayed a lower IgM concentration than samples stored for shorter periods. And in 1978 Guimarães et al. (15) showed by radial immunodiffusion tests that eluates stored for 2.5 months tended to have lower IgG concentrations than those found before storage.

In the present article we have analyzed the effects of long-term storage at -20°C upon the antibody combining sites of IgG and IgM (as indicated by immunofluorescence tests) and upon class-specific determinants (as indicated by radial immunodiffusion tests) using antibody molecules from blood stored on filter paper and from sera that were stored frozen after addition of glycerin, freeze-drying, or no treatment.

Materials and Methods

Thirty milliliters of venous blood were withdrawn from each of 23 patients with clinical diagnoses of toxoplasmosis and previous se-

rology indicating that they had specific IgM-anti-*Toxoplasma gondii* antibodies (16).⁷ Immediately after withdrawal, about 10ml of each blood sample were spread evenly over a $10 \times 7.5\text{cm}$ Whatman No. 1 filter paper so as to cover the whole paper strip with a thin but unbroken layer of blood. This was done by removing the needle from the tip of the syringe and allowing blood to flow out by gravity. The tip was moved steadily along, spreading an uninterrupted blood layer over the whole paper.

The filter-paper samples were then left to dry at room temperature for 60 minutes. After that they were placed in plastic bags that were folded and stored inside screw-topped glass containers at -20°C throughout the study.

For testing purposes, 4cm^2 circles were punched out of the papers and eluted with 0.25ml of 0.01M phosphate-buffered saline (PBS) for two hours at 4°C . According to Souza and Camargo (10), this results in the equivalent of a 1:5 dilution. The remaining blood was left to clot at room temperature.

Regarding the remaining volume of each blood sample (about 20ml), following clot retraction two-thirds of the resulting serum was placed in glass ampoules, 0.5ml being placed in each ampoule. In addition, half the ampoules received 0.5ml of neutral analytical grade glycerin (Merck). All the ampoules were sealed with a Bunsen burner. The remaining third of each serum sample was then divided into 0.5ml aliquots and freeze-dried for 18 hours at 0.05 Tor. Before testing they were reconstituted with distilled water to their original 0.5ml volume. All the aliquots were kept at -20°C until tested. The tests em-

⁷ These patients were referred to one of us (MCSG) by Professor Amato from the inpatient clinic at the Department of Tropical Medicine and Dermatology (University of São Paulo Medical School) and from his private practice. The patients, of whom 11 were male and 12 female, ranged in age from nine to 58 years. The blood samples were drawn by MCSG at the department's seroepidemiology laboratory. Sample collection began in March 1977 and ended in March 1980.

ployed (immunofluorescence and radial immunodiffusion) were performed on aliquots of all the samples after 0, 3, 6, 9, 12, and 18 months of storage.

Immunofluorescence (IF) Tests

Immunofluorescence (IF) testing was performed according to the method described by Camargo (17), using formalin-fixed *Toxoplasma gondii* as antigen and FITC-antihuman IgG and FITC-antihuman IgM conjugates.⁸ The conjugates were prepared and used according to the method of Camargo et al. (18) from antisera produced at the Institute of Tropical Medicine of São Paulo. Both of the conjugates had a F/P (fluorescein to protein) ratio equal to 3. The *T. gondii* antigen was also prepared according to the method of Camargo et al. (18).

Aliquots of all the samples (reconstituted freeze-dried sera, frozen sera, sera preserved in glycerin, and filter-paper eluates) were diluted with 0.15M NaCl. For all but the sera preserved in glycerin the starting IgG-immunofluorescence dilution was 1:1,024 and the starting IgM-immunofluorescence dilution was 1:64. In the case of the sera preserved in glycerin, the amount of 0.15M NaCl added was half of that added to the other sera. The titer end-point recorded was the last dilution that produced an uninterrupted, bright apple-green fluorescence on the parasite's membrane. Polar parasite staining was considered a negative result.

This test system's reproducibility was assessed by including positive and negative standard sera. These were titrated with the other sera and on each occasion were found to yield unvarying results.

Radial Immunodiffusion (RID) Tests

IgG and IgM radial immunodiffusion tests were performed as described by Guimarães et

al. (15). All the samples tested were tested in duplicate, and the results were averaged for purposes of immunoglobulin measurement. The samples' IgG and IgM protein concentrations were assessed on the basis of standard curves constructed from IgG standards of 43, 100, and 191 mg/ml and IgM standards of 63, 125, and 261 mg/ml, respectively. The IgG and IgM standards were the same used for "Tripartigen" radial immunodiffusion tests (Behringwerke AG, Marburg, West Germany).

Statistical Analyses

For statistical purposes, the IF serum titers were transformed from the geometric form to the arithmetic form ($\log_{10} (x + 1)$).

In addition, a multivariate general linear model was used to analyze the IgG and IgM levels found by means of IF or RID after 3, 6, 9, 12, and 18 months of storage. For hypotheses testing purposes, the Wilks statistics were used according to the Roy and Bose method (19, 20). Multiple comparison was accomplished by the Newman-Keuls procedure (21). Titer differences for the 0-3 month interval were evaluated by assessment of 95% confidence interval limits. Whenever 95% confidence interval limits were assessed, if they were found to be symmetrically distributed around zero the results were considered nonsignificant.

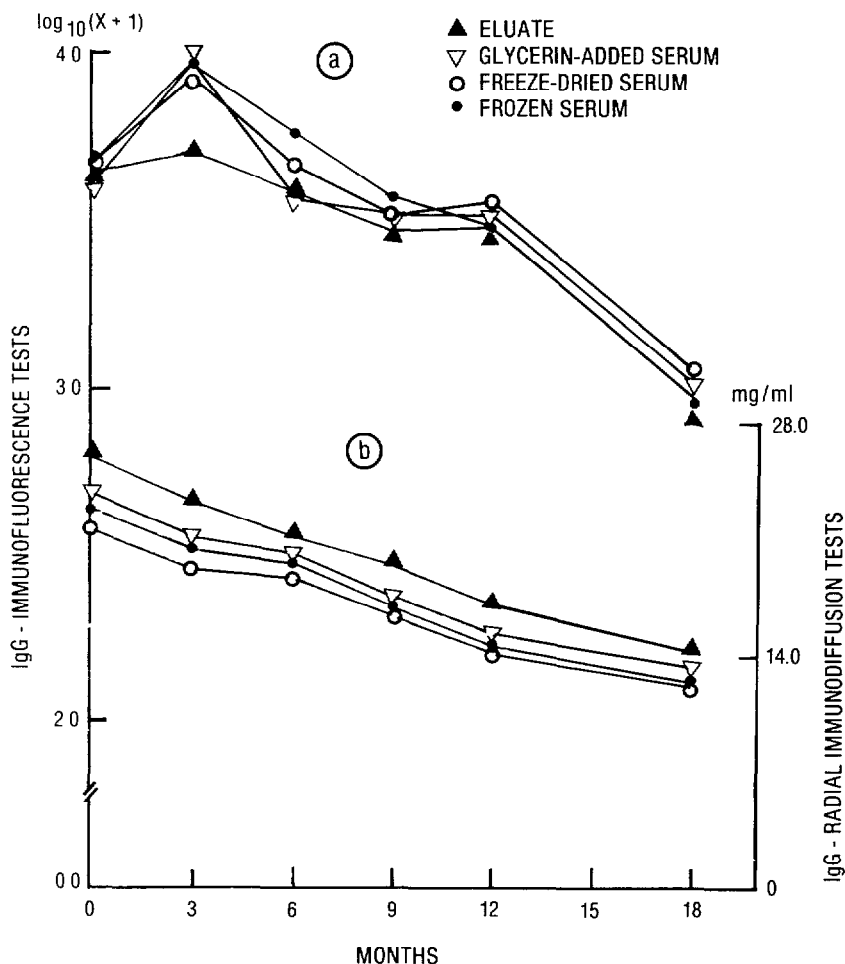
Results

IgG Findings

Immunofluorescence (IF) tests. The IF tests of the sera stored in different ways yielded geometric mean titers (GMT) indicating no significant difference between the four sets of sera before storage began (at time zero). GMT after three months of storage also showed no significant differences for the 0-3 month interval (Figure 1a), as indicated by 95% confidence interval limits.

⁸ FITC = fluorescein isothiocyanate.

Figure 1. IgG data: IF and RID test results for IgG in the filter-paper eluates and sera stored three ways, by length of storage time.



In contrast, there did appear to be statistically significant differences between these early GMTs and those obtained later, regardless of the storage method used. Overall, p was 0.0161 for 3–6 months, 0.0378 for 3–9 months, 0.0201 for 3–12 months, and 0.0006 for 3–18 months.

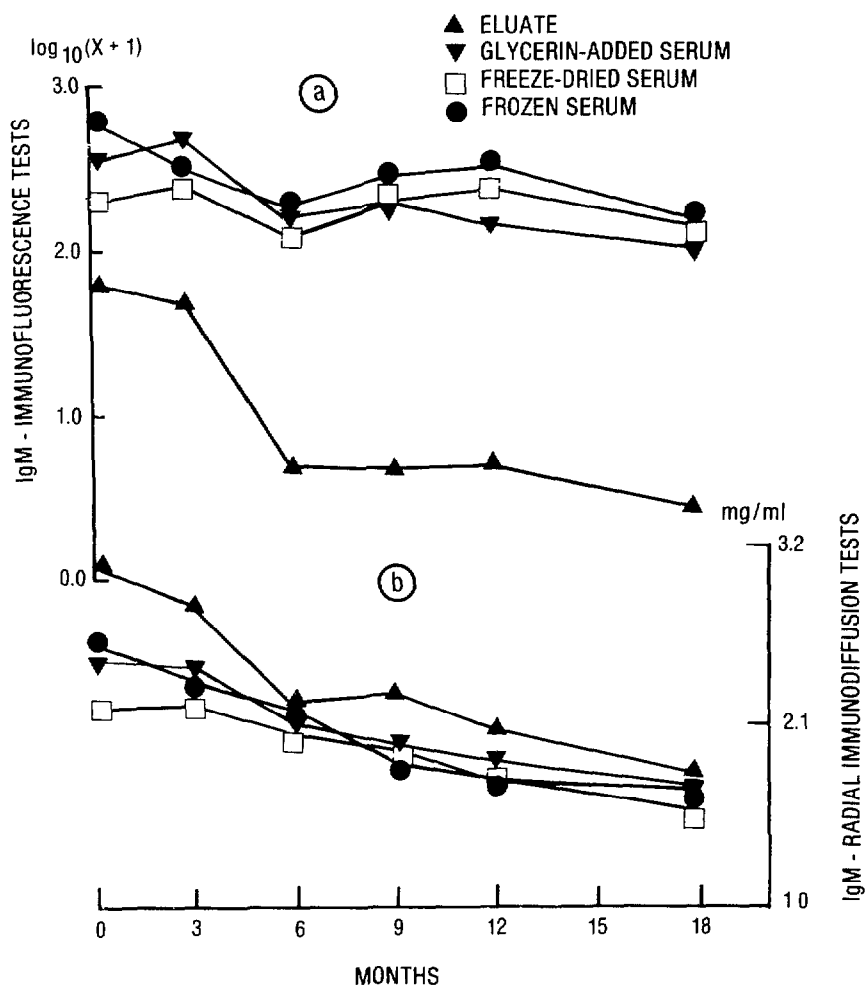
Radial immunodiffusion (RID) tests. The RID tests found mean IgG antibody concentrations in the eluate samples at time zero that were significantly higher than the mean concentrations found in the samples stored in other ways (Figure 1b). They also found that

mean IgG concentrations in each set of samples stored a particular way were higher at time zero than they were after three months of storage (see the 95% confidence interval limits for the 0–3 month period shown in Table 1). Similarly, for any given storage procedure the mean IgG concentration at three months was significantly higher than the mean concentrations at nine, 12, and 18 months, irrespective of the storage procedure used. However, significant differences were not found for all storage procedures combined with respect to the three-month and six-month data.

Table 1. Ninety-five per cent confidence interval limits for 0-3 month storage of IgG and IgM by the four study methods, as indicated by radial immunodiffusion (RID) test results.

Material tested	95% confidence intervals for IgG	95% confidence intervals for IgM
Blood stored on filter paper (eluate)	[1.209; 4.461]	[-0.175; 0.731]
Sera with glycerin added	[0.616; 2.419]	[-0.155; 0.355]
Freeze-dried sera	[1.146; 3.054]	[-0.268; 0.215]
Frozen sera	[0.645; 4.034]	[-0.160; 0.516]

Figure 2. IgM data: IF and RID test results for IgM in the filter-paper eluates and sera stored three ways, by length of storage time.



Overall, the mean IgG concentration found in the filter-paper eluate was significantly higher at any given time (see Figure 1b) than the mean concentrations found in samples stored by any other method. (The value of p for the filter-paper eluate versus the glycerin-added sera was 0.0033, for the eluate versus the freeze-dried sera was 0.001, and for the eluate versus the frozen sera was 0.0006.)

IgM Findings

Immunofluorescence (IF) tests. The GMT obtained with the filter-paper eluates at time zero was significantly lower than the GMTs obtained with the samples stored by other methods. This difference was still significant after three months of storage ($p < 0.0001$), and over the 3–6 month period there was a sharp decline in the eluate GMT that widened the gap considerably for the rest of the study period (Figure 2a). The statistically significant difference between the eluate GMTs at three months and at six months is demonstrated by the 95% confidence interval limits shown in Table 2.

Radial immunodiffusion (RID) tests. Figure 2b shows the mean IgM concentrations found by RID testing for each storage procedure throughout the observation period. These data indicate that at time zero the filter-paper eluates' mean IgM concentration was significantly higher than the mean IgM concentration in the samples stored by any other method (see the 95% confidence interval limits shown in Table 1). Similarly, the

eluates' mean IgM concentration was found to be higher than those of the samples stored by other means over the 0–3 month period. However, during the 6–18 month interval the eluates' mean IgM concentrations were found not to differ significantly from the mean concentrations in samples stored by other means. For samples stored in any given way, the mean IgM concentrations were found to be significantly lower at 12 months and at 18 months than at three months, irrespective of the storage procedure used ($p = 0.0002$ and $p < 0.001$, respectively).

Discussion

The foregoing results indicate that IgG and IgM denaturation over time (expressed as a decline in IF test GMTs and in RID test mean concentrations) took place in different parts of the molecules studied, depending on whether IgG or IgM was being tested.

As stated previously, it was initially assumed for testing purposes that the serum dilution represented by the eluate was 1:5 (10). However, as shown by Guimarães et al. (15), previous attempts to corroborate this point have proven unsuccessful. This is relevant in view of the fact that the eluates' mean IgG and IgM concentrations, as determined by RID testing (Tables 3 and 4), were consistently higher than the mean concentrations in the three sets of samples stored in other ways throughout the observation period. This difference is attributed to an error in assessing the eluate dilution.

Table 2. Ninety-five per cent confidence interval limits for 0–3 month storage of IgG and IgM by the four study methods, as indicated by immunofluorescence (IF) test results.

Material tested	95% confidence intervals for IgG	95% confidence intervals for IgM
Blood stored on filter paper (eluate)	[−0.602; 0.472]	[−0.667; 0.931]
Sera with glycerin added	[−0.780; 0.048]	[−0.476; 0.370]
Freeze-dried sera	[−0.688; 0.164]	[−0.706; 0.410]
Frozen sera	[−0.742; 0.114]	[−0.102; 0.785]

Table 3. Mean IgG concentrations, one standard deviation (S.D.) from each mean, and median IgG concentrations in samples stored by the four study methods after 0, 3, 6, 9, 12, and 18 months of storage, as indicated by RID test results.

Material tested		Reading after indicated months of storage, in mg per ml					
		0 months	3 months	6 months	9 months	12 months	18 months
Blood stored on filter paper (eluate)	Mean	26.26	23.42	21.47	19.70	16.58	14.42
	S.D.	±5.51	±5.45	±4.98	±4.38	±3.29	±3.29
	Median	25.6	22.0	20.6	19.2	15.9	14.2
Sera with glycerin added	Mean	22.80	21.29	20.34	17.66	15.56	13.55
	S.D.	±5.17	±5.08	±5.06	±4.27	±3.57	±3.41
	Median	20.6	21.2	19.2	16.4	16.8	13.2
Freeze-dried sera	Mean	21.63	19.40	18.81	16.58	14.50	12.33
	S.D.	±4.62	±4.47	±4.86	±4.50	±4.32	±4.53
	Median	20.4	18.0	16.9	15.8	14.7	11.2
Frozen sera	Mean	23.04	20.66	19.73	17.13	14.92	12.79
	S.D.	±5.18	±5.40	±4.75	±4.40	±3.58	±3.84
	Median	21.9	19.0	20.6	16.1	15.6	12.6

Table 4. Mean IgM concentrations, one standard deviation (S.D.) from each mean, and median IgM concentrations in samples stored by the four study methods after 0, 3, 6, 9, 12, and 18 months of storage, as indicated by RID test results.

Material tested		Reading after indicated months of storage, in mg per ml					
		0 months	3 months	6 months	9 months	12 months	18 months
Blood stored on filter paper (eluate)	Mean	3.10	2.83	2.20	2.30	2.07	1.79
	S.D.	±2.38	±1.93	±1.49	±1.60	±1.49	±1.14
	Median	2.0	2.1	1.8	1.9	1.5	1.4
Sera with glycerin added	Mean	2.52	2.44	2.16	1.97	1.88	1.73
	S.D.	±1.73	±1.57	±1.27	±1.35	±1.30	±1.19
	Median	2.0	2.1	1.8	1.9	1.5	1.4
Freeze-dried sera	Mean	2.20	2.22	2.05	1.93	1.76	1.59
	S.D.	±1.34	±1.36	±1.21	±1.53	±1.37	±1.10
	Median	2.0	1.6	2.0	1.5	1.3	1.1
Frozen sera	Mean	2.59	2.41	2.19	1.91	1.78	1.69
	S.D.	±1.82	±1.58	±1.29	±1.21	±1.16	±1.16
	Median	2.4	2.0	1.8	1.6	1.4	1.2

Over time, the samples not stored on filter paper showed IgG and IgM concentrations that were consistently lower than their initial concentrations. Nevertheless, when the eluate IgG GMTs are taken as a reference (see Table 5), except for glycerin-added sera at time zero and frozen sera at times 12 and 18 months, all other storage procedures yielded higher GMT values. In the case of IgM GMTs (see Table 6), this difference was even more marked and became particularly pronounced during the 3–6 month period. Moreover, while the differences between eluate GMTs and glycerin-added serum GMTs were not statistically significant at zero and six months for IgG, their IgM values (as previously noted in the Results section) were significantly different.

Since the time when filter paper was proposed as a convenient medium for blood collection and storage, authors have mentioned difficulties in estimating the precise amount of blood placed on the paper. Errors in making this estimation logically translate into differences between serum and eluate end-point titrations.

The discrepancies reported here remained unrecognized for so long because the IF conjugates employed were mainly anti-gammaglobulin conjugates rather than anti-immunoglobulin conjugates; and since the IgG concentration in serum is some 15 times greater than the IgM concentration, any reduction in eluted IgM values tended to be masked by the remaining IgG molecules bearing antibody-combining sites complementary to the same antigenic determinants (7–10, 22).

Recently, Roffi et al. (23) reported that a correct assessment of the serum dilution represented by the eluate could be made by evaluating blood samples' hemoglobin content (by cyanometahemoglobin development) and red blood cell sedimentation rates. In 17 such determinations made by the authors, the average dilution was found to be 1:528, with a titer range of 1:370 to 1:700. At present there are still no simple ways to guarantee that the eluate will correspond to a given serum dilu-

tion, except by comparing serum and eluate titers and immunoglobulin concentrations—as was done in the present study, where RID test results showed mean IgG concentrations to be higher in the eluate than in the sera stored by other means, both at time zero and throughout the observation period (see Table 3 and Figure 1b).

IgG Antibodies

IgG antibodies detected by IF testing had GMTs at 18 months that were significantly lower than those found at three months, irrespective of the storage procedure used. GMT declines registered at 12 months, while noteworthy, were not so marked (see Figure 1a). Thus, regarding blood samples collected and stored on filter paper for seroepidemiologic purposes, it would appear that such samples may be kept for a maximum storage period of 12 months under the conditions described here without loss of confidence in the results. This is an important point to consider in large-scale surveys, where samples must be stored for several months before being processed.

The RID testing (Figure 1b) found mean IgG concentrations to be significantly lower at nine months than at zero months, irrespective of the storage procedure.

IgM Antibodies

There was a sharp contrast in the IF test results between the eluate GMTs and the GMTs obtained with sera stored using the three other procedures. From time zero onward, and especially from six months onward, the eluate GMTs for IgM antibodies were markedly lower than the others (see Figure 2b), a difference that was statistically significant. More specifically, at time zero eluates from only two of the 23 samples (8.7%) yielded negative IF results for IgM. But at three months eluates from seven samples

Table 5. Geometric mean IgG titers (GMT), arithmetic mean IgG titers,^a one standard deviation (S.D.) from each arithmetic mean, and the IgG titer ranges found in samples stored by the four study methods after 0, 3, 6, 9, 12, and 18 months of storage, as indicated by IF test results.

Material tested		Titers, standard deviations, and ranges after indicated months of storage					
		0 months	3 months	6 months	9 months	12 months	18 months
Blood stored on filter paper (eluate)	GMT	4,481.17	5,210.38	3,973.11	3,029.03	3,121.70	964.12
	Arithmetic mean	3.651 ^a	3.717	3.599	3.481	3.494	2.984
	S.D.	±0.92 ^a	±0.96	±0.36	±0.83	±0.85	±1.22
	Titer range	4.816 ^a	4.515	0.902	4.214	4.214	3.913
Sera with glycerin added	GMT	3,854.78	8,960.82	3,521.24	3,315.95	3,315.95	1,087.40
	Arithmetic mean	3.586	3.952	3.547	3.521	3.521	3.036
	S.D.	±0.90	±0.46	±0.86	±0.87	±0.88	±1.23
	Titer range	4.816	1.805	4.515	4.214	4.515	3.913
Freeze-dried sera	GMT	4,753.35	8,696.57	4,760.97	3,417.06	3,628.96	1,190.88
	Arithmetic mean	3.677	3.939	3.678	3.534	3.560	3.076
	S.D.	±0.91	±0.56	±0.47	±0.86	±0.88	±1.03
	Titer range	5.117	1.805	1.504	4.214	4.816	3.913
Frozen sera	GMT	4,618.29	9,516.52	5,877.25	3,854.01	3,121.70	964.12
	Arithmetic mean	3.664	3.976	3.769	3.586	3.494	2.984
	S.D.	±0.92	±0.46	±0.41	±0.85	±0.86	±1.22
	Titer range	4.816	1.805	1.504	4.214	4.816	3.913

^alog₁₀ (x + 1).

Table 6. Geometric mean IgM titers (GMT), arithmetic mean IgM titers,^a one standard deviation (S.D.) from each arithmetic mean, and the IgM titer ranges found in samples stored by the four study methods after 0, 3, 6, 9, 12, and 18 months of storage, as indicated by IF test results.

Material tested		Titers, standard deviations, and ranges after indicated months of storage					
		0 months	3 months	6 months	9 months	12 months	18 months
Blood stored on filter paper (eluate)	GMT	66.25	48.99	5.10	4.80	5.09	2.79
	Arithmetic mean	1.821 ^a	1.699	0.708	0.681	0.707	0.445
	S.D.	±1.38 ^a	±1.64	±1.13	±1.22	±1.23	±0.99
	Titer range	4.214 ^a	4.515	3.011	3.612	3.011	3.011
Sera with glycerin added	GMT	368.64	469.28	153.74	196.04	149.40	104.16
	Arithmetic mean	2.567	2.671	2.187	2.292	2.174	2.018
	S.D.	±1.22	±1.11	±1.26	±1.06	±1.16	±1.02
	Titer range	4.214	4.124	3.913	3.913	4.214	3.011
Freeze-dried sera	GMT	207.82	256.45	117.41	208.14	242.05	140.80
	Arithmetic mean	2.318	2.409	2.070	2.318	2.384	2.149
	S.D.	±1.40	±1.43	±1.21	±1.06	±0.93	±0.94
	Titer range	4.214	4.214	3.612	3.913	4.214	3.011
Frozen sera	GMT	634.95	317.08	179.02	298.76	327.21	158.74
	Arithmetic mean	2.803	2.501	2.253	2.475	2.514	2.201
	S.D.	±0.75	±1.22	±1.22	±0.96	±0.80	±0.95
	Titer range	2.401	4.214	3.913	3.913	4.214	3.011

^alog₁₀ (x + 1).

(30.4%) were negative, at six months 10 (43.5%) were negative, and at 18 months only four (17.4%) were positive.

There could be advantages to this. For if the finding is correct and accurate detection of IgM antibodies is in fact limited to samples that have been stored on filter paper for not more than 90 days, that circumstance should also permit researchers to evaluate IgG antibodies for the remainder of the acceptable IgG storage period with sharply reduced interference from IgM antibodies—which can be a cause of nonspecific results in parasitic disease serology (16, 24).

In contrast to these IF results, RID testing for IgM indicated that the eluate's mean IgM concentration was higher than the mean IgM concentration found in the freeze-dried sera, frozen sera, or glycerin-added sera at time zero. Irrespective of the storage procedure, however, the test results showed a progressive decline in mean IgM concentrations over time that prevailed throughout the 18-month study period and became statistically significant at 12 months.

Concluding Remarks

Despite the limitations already described regarding the maximum length of antibody storage (especially IgM antibody storage) by the filter-paper method, and despite the still unsolved problem of how to determine what serum dilution is represented by the filter-paper eluate, we believe that for seroepidemiologic purposes this is the best method avail-

able for collecting and storing blood samples. Since only a small amount of blood is withdrawn, usually from the subject's fingertip, the procedure permits collection of samples from all age groups, including newborns and the elderly; it dispenses with the need for syringes, test tubes, strictly sterile conditions, and refrigeration; it keeps transport costs to a minimum; and it provides specimens suitable for many serologic tests—including immunofluorescence, ELISA, complement fixation (for tests where the end-point titration is assessed by the bulk of unlysed red blood cells deposited at the bottom of either a test tube or a plastic plate), and hemagglutination. Also, since collection of blood by finger-prick on filter paper does not require skilled personnel, this procedure permits collection of such samples by community leaders or community health workers.

Validation of this procedure was accomplished in Brazil by a nationwide seroepidemiologic survey of Chagas' disease in which approximately 1,600,000 blood samples from rural areas were collected on filter paper and screened for anti-*T. cruzi* antibodies by IF tests employing an anti-IgG conjugate. Duplicate testing indicated a loss of approximately 10% of the samples as a result of improper labeling or fungus contamination (M. E. Camargo, personal communication). Although the samples were tested between two and three months after collection in this case, it could well be that further delay would have improved the tests' specificity, as a result of the previously mentioned denaturation of IgM antibodies.

SUMMARY

To help determine how long IgG and IgM antibodies can be stored in blood samples collected on filter paper, blood samples were obtained from 23 toxoplasmosis patients. A portion of each sample was then stored on filter paper at -20°C , and serum from the remainder of the sample was divided into thirds—one-third being untreated, one-third

being combined with an equal volume of glycerin, and one-third being freeze-dried—before being stored at -20°C . Portions of each sample stored in these four ways were then examined for IgG and IgM antibodies by immunofluorescence (IF) and radial immunodiffusion (RID) tests after having been stored for 0, 3, 6, 9, 12, and 18 months.

The results illustrated the difficulty of estimating what serum dilution is equivalent to the eluate obtained from the filter-paper specimens for testing purposes. They also indicated that deterioration of IgG occurred faster in the samples stored on filter paper than it did in the samples stored in other ways. This relatively faster deterioration of the filter-paper samples was even more marked with regard to IgM.

Despite these limitations, however, it is suggested that the procedure of collecting blood samples by finger-prick and storing them on filter pa-

per is still the best available for seroepidemiologic purposes—because it requires only a small amount of blood; because it does not require syringes, test tubes, strictly sterile conditions, or refrigeration; because it provides specimens suitable for many serologic tests; and because it can be performed by relatively unskilled personnel. Also, the relatively fast deterioration of IgM observed in this study suggests it might be possible to reduce IgM interference in testing for IgG antibodies by storing the test samples long enough for such IgM deterioration to occur.

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NEW CHAIRMAN FOR CARIBBEAN RESEARCH COUNCIL

Sir Arthur Lewis, currently the James Madison Professor of Political Economy at Princeton University, has accepted an invitation to be Chairman of the Commonwealth Caribbean Medical Research Council (CCMRC). The Council, which is the only regional medical research organization in the Caribbean, advises the Caribbean Community's Conference of Ministers Responsible for Health about the priorities and conduct of medical research. Its annual scientific meetings attract widespread support, and many studies of major importance to the region have received their first airing at these meetings.

Sir Arthur, who was born in Saint Lucia and who has had an outstanding career as a scholar and public servant, served as Vice-Chancellor of the University of the West Indies from 1959 to 1963 and subsequently as the Chancellor of the University of Guyana. A frequent adviser to governments in the region, Sir Arthur has received over 30 honorary degrees from universities. He was created a Knight Bachelor in 1963 and received the Nobel Prize in Economics in 1979.

The new CCMRC Chairman succeeds Sir Hugh Springer, who was Chairman from 1965 until his appointment as Governor General of Barbados in 1984.