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a minimum of handling, using disposable gloves, in plastic bags. The plastic bags should be heat-sealed and sent in suitable containers to the forensic laboratory for further examination by standard "no-touch" microbiological techniques.

The wearing of respirators by police officers in the field when handling human biological fluids and products is unnecessary. The use of protective gowns will be dictated by special circumstances such as massive contamination of a body with blood.

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TESTING OF PLASMA POOLS FOR HEPATITIS B SURFACE ANTIGEN BY THIRD GENERATION TESTS DURING 1975 AND 1976

B.S. Combridge and W.d'A. Maycock,
Blood Products Laboratory,
Lister Institute,
Elstree, Hertfordshire, England.

Blood Products Laboratory (BPL) received from Regional Transfusion Centres, (RTCs) 10- and 25- donor plasma pools composed either of fresh-frozen plasma or plasma from time-expired blood. All individual donations were screened at RTCs by reversed passive haemagglutination (RPH) ("Hepatest"). One RTC used counter immunoelectrophoresis (CIE) for period Jan/June 1975.

In 1975 all pools were tested at BPL by RPH and positives were retested by RIA ("Ausria II"). The results are shown in Table 1.

Table 1.

(1) Pools Tested	(2) Antigen positive (RPH)	(3) Confirmatory RIA tests (Prof. A.J. Zuckerman)
22,737	34	25 positive and neutralized 9 positive and "diluted" during neutralization

RTCs retested donors who had contributed to the 34 positive pools. To date 12 HBsAg positive donors have been detected one of whom contributed to each of 12 pools among the 25 RIA confirmed antigen-positive pools and two among contributors to two of the 9 "diluted out" pools.

In 1976 all pools were tested at BPL by "Ausria II" and individual donations screened by RPH at RTCs. All antigen positive pools of time-expired and all pools of fresh-frozen plasma were also tested by a modified RIA method (Dr. Dane, Medical School, Middlesex Hospital, London, W.1.), intermediate in sensitivity between "Ausria II" and the method described by Cameron & Dane (Brit. med. Bull. 1974, 30, 90). The total number of pools tested was 23,283. The results are shown in Table 2.

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Table 2.
Antigen Positive ("Ausria II").

	(1) Pools	(2) Pos/neg ratio > 2·1	(3) Pos/neg ratio 1·5 -2·1	(4) Positive Modified RIA method. All pos/neg ratio > 2·1
Time-expired plasma	17,693	25	3	28
Fresh frozen plasma	5,590	8	2	11

The modified RIA method detected one antigen positive pool undetected by "Ausria II".

During follow-up an antigen-positive donor was detected by RIA in each of 4 of the 5 borderline positive pools (col.3). All four donors were RPH negative.

All the "Ausria II" positives (col.2) were confirmed by Professor A.J. Zuckerman using "Ausria II". The positives (cols.2 & 3) together with the positive detected only by the modified RIA method (col.4) were confirmed by a more sensitive test (Drs. Cameron and Dane).

The experience at BPL suggests that pools of plasma, especially if they are to be used to prepare fibrinogen products, should be screened, preferably by a radioimmunoassay method, even though the individual donations composing the pools have already been screened. The observations in 1975 suggest that this should be done even though the methods used to screen donors and pools are the same. One possible explanation of the failure to detect antigen in some donors is that the titre was subthreshold when the donation used for pooling was tested.

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Association of e antigen with Dane particle DNA in sera from asymptomatic carriers of hepatitis B surface antigen.

Barbara G. Werner, Anna P. O'Connell and Jesse Summers

The Institute for Cancer Research, The Fox Chase Cancer Center, 7701 Burholme Avenue, Philadelphia, Pennsylvania 19111

Sera containing hepatitis B surface antigen from thirty asymptomatic blood donors were assayed for e antigen (HBeAg) and antibody to e antigen (anti-HBe) by rheophoresis. Fourteen (47%) had detectable HBeAg, ten (33%) had anti-HBe and six donors (20%) had neither.

DNA was extracted from 26 of these sera and assayed for its ability to anneal to a ^{32}P DNA probe that is a copy of Dane particle DNA. All 10 HBeAg positive samples tested contained DNA that formed specific hybrids with the DNA probe as did one of 10 anti-HBe positive samples. Hybridization was not detected using nine sera containing anti-HBe and six sera without HBeAg or anti-HBe.

Since the Dane particle is thought to be the hepatitis B virus, this association between HBeAg positivity and Dane particle

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DNA strongly supports the hypothesis that e antigen is a marker of the presence of the virus and, consequently, potential infectivity. Sera with anti-HBe, on the other hand, seem much less likely to be infective since the probability of finding viral DNA in these sera is greatly reduced, and when present, the levels are lower.

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T-LYMPHOCYTES IN MARMOSETS WITH NON-B HEPATITIS

D. R. Johnson, D. A. Peterson, L. G. Wolfe and F. W. Deinhardt

Departments of Microbiology, Rush-Presbyterian-St. Luke's and University of Illinois Medical Centers, Chicago, Illinois 60612

During hepatitis B infection in man certain functions of the T-lymphocyte population are depressed, perhaps contributing to persistence of viral replication and development of a chronic carrier state. To determine the effect of viral hepatitis on the T-lymphocyte population of marmosets, we have measured the percentages of circulating lymphocytes forming rosettes with sheep erythrocytes (E-rosettes) in marmosets experimentally infected with non-B human hepatitis. Fourteen adult marmosets (Saguinus fuscicollis, S. nigricollis) were divided into 3 groups: (a) 4 uninoculated control animals (b) 6 marmosets inoculated intramuscularly with 0.5 ml of a 5% (w/v) human fecal suspension containing the MS-1 prototype hepatitis A virus (c) 4 animals inoculated intramuscularly with 0.5 ml of acute phase marmoset serum GB strain, passage 11 (non A - non B human hepatitis) diluted 1:50 in HBSS. The marmosets were evaluated for plasma transaminase levels (SGOT/SGPT) weekly, liver biopsies at two week intervals for liver histology and percentages of circulating lymphocytes forming E-rosettes were determined at weekly intervals.

Marmosets in group b (prototype strain of hepatitis A [MS-1]) developed slight increases in serum transaminase levels with negligible histopathologic lesions although 4 of 6 developed anti-hepatitis A antibodies (anti-HA) demonstrable by immune adherence tests. Marmosets in group c (GB strain hepatitis) developed enzymatic and morphologic evidence of hepatitis 14-21 days post-inoculation which persisted for 2-3 weeks.

Marmosets inoculated with the GB strain showed a significant decrease ($p<.001$) of E-rosette forming cells slightly before and during the acute phase of the disease (days 14 through 28 post-inoculation); E-rosette-forming cells decreased from 60-65% in pre-inoculation samples to 35-40% during the acute phase of the disease and then returned to baseline values by 5 weeks post-inoculation. Marmosets inoculated with MS-1 developed less severe, but significant decreases ($p<.001$) of E-rosette forming cells during a period which correlated with slight elevations of serum transaminase levels. E-rosette forming cells decreased from normal levels to 40-55% in the 4 animals which developed anti-HAV and decreased to only 55-60% in the two animals which did not seroconvert. The number of circulating E-rosette forming cells in control animals remained within normal ranges (58-67%) throughout the observation period.

Chisari et al., (J. Exptl. Med. 142:1092, 1975) have reported the presence of a serum lipoprotein called rosette inhibitory factor (RIF) in human hepatitis B patients which inhibits E-rosette formation in vitro. Plasma from a marmoset infected with the GB strain of hepatitis was examined for the presence of RIF. The drop (65-35%) in circulating E-rosette forming cells observed during the acute phase of the disease (14-28 days post-inoculation) correlated with enhanced activity of a plasma factor that inhibited E-rosette formation by normal marmoset lymphocytes. This effect was reversible, as RIF treated lymphocytes formed E-rosettes normally when they were washed, incubated overnight at 37°C and again washed before the E-rosette test. These results are similar to findings in humans with hepatitis B and alcoholic liver disease. The plasma RIF probably accounts for part but

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not all of the inhibition of E-rosette formation by T lymphocytes during the acute phase of viral hepatitis experimentally induced in marmosets.

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PRACTICAL GUIDELINES FOR ASSESSING PATIENTS POSITIVE FOR HEPATITIS B SURFACE ANTIGEN

S. V. Feinman, B. Berris, J. C. Sinclair and D. M. Wrobel

The Liver Clinic, Department of Medicine, Mount Sinai Hospital, University of Toronto and the Toronto Red Cross Centre.

In assessing the patient with hepatitis B surface antigen (HB_sAg), the physician must decide on the basis of physical findings, results of laboratory tests and biopsy, when indicated, whether the patient is an asymptomatic carrier or has acute or chronic hepatitis. (See Tables I and II.) Asymptomatic carriers of HB_sAg must be educated in personal hygiene and the possibility of transmission. They should not be allowed to donate blood or breast-feed and they should not work with blood products for human use or pharmaceutical products designated for intravenous use. However, it is otherwise not necessary to advise these individuals to change their profession.

TABLE I - FACTORS IN THE ACQUISITION OF HEPATITIS B SURFACE ANTIGEN (HB_sAg)

FACTOR	COMMENT
Proven Parenteral exposure to hepatitis B virus, resulting in hepatitis (icteric or non-icteric)	After having hepatitis B, 10% of patients remain HB_sAg positive; of these, 5% have chronic hepatitis and 5% are asymptomatic.
Subclinical hepatitis B without proven parenteral exposure	Of 115 carriers only 1 gave a history of icteric hepatitis. Common in Southeast Asia, Mediterranean area and Africa. Imporatnce of low-dose infections via saliva or other biologic fluids unknown.
Maternal exposure	
Introduction of HB_sAg in clusters by migration from one country to another	In Ontario, 70% of cases occur in immigrants from Mediterranean area and Far East.

TABLE II - PHYSICAL & LABORATORY ASSESSMENT OF HB_sAg POSITIVE INDIVIDUALS

HB_sAg POSITIVE GROUP	PHYSICAL FINDINGS	LIVER FUNCTION	CONCLUSION
ASYMPTOMATIC	Normal	Normal	Asymptomatic carrier of HB_sAg
	Normal	Abnormal	Incubation period of acute hepatitis B
	Abnormal	Abnormal	Chronic hepatitis with antigenemia
SYMPTOMATIC	Normal	Normal	Prodromal or resolving hepatitis B
	Normal	Abnormal	Prodromal or resolving hepatitis B
	Abnormal	Abnormal	Hepatitis B, acute or chronic or cirrhotic