AUSTRALIA ANTIGEN - PRESENT STATUS

Parkland Memorial Hospital

Medical Grand Rounds

January 6, 1972

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Historical Background

During the early 1960's Blumberg and associates were engaged in studies of genetic variations of human serum lipoprotein antigens for use as genetic markers in population studies (1). The Ouchterlony technique was used in which test sera and presumptive antisera were placed in separate wells on agar-gel coated slides. After incubation, the slides were inspected for precipitin lines indicating an antigen-antibody reaction between adjacent wells. The result of this effort was the description of the "Ag" low density lipoprotein antigen system.

When extended studies were undertaken, screening sera from several different geographical areas of the world, a new precipitin line indicating an antigen distinct from the Ag system was obtained with the serum of an Australian aborigine. In view of its uncertain nature, this reactant was assigned the noncommittal name "Australia antigen".* (2).

Between 1967 and 1969 Blumberg's group examined sera of several hundred patients for the presence of Au antigen. The first group noted to have a high prevalence of Au was leukemia patients (2) and the possibility was considered that Au was related to a leukemogenic virus. Nevertheless, the favored theory was that Au, like the earlier Ag system, represented a genetically determined antigenic variant of a serum lipoprotein. Indeed, when it was realized that Au was present in large numbers of persons in certain tropical populations, family studies in such areas (3, 4) revealed a pattern very suggestive of an autosomal recessive mode of inheritance of the Au-"trait". The high incidence of Au in certain patient groups in the United States, including persons with Down's syndrome (mongolism), chronic renal failure and various forms of leukemia was difficult to reconcile with the genetic postulate.

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* Blumberg's group subsequently described another antigen which they felt was related to Australia antigen and assigned terms Au (1) and Au (2) to the original and the "new" antigen respectively (5). No further confirmatory data have appeared regarding Au(2) and its significance is in doubt. Nevertheless, publications from that laboratory continue to refer to Au(1). Other terms applied to Australia antigen include Hepatitis-associated antigen (HAA), Hepatitis Antigen (HA), SH antigen and Au/SH. Since Au is also associated with conditions apart from viral hepatitis, these terms appear to have no advantage over the generally understood term "Australia antigen".)

The first suggestion that Au might be a hepatitis virus, or related to such a virus, was made by Blumberg in 1967 (6). As confirmatory data began to accumulate (7, 8), the new concept evolved that Au was, in fact, related to an infectious agent, commonly but not exclusively transmitted by the parenteral route (especially by blood transfusion) causing acute hepatitis in some persons, but persisting as an apparently innocuous carrier state in some genetically predisposed individuals (3) as well as in patients with impaired resistance mechanisms - the leukemics, mongoloids and chronic renal failure patients previously mentioned.

VIRAL STATUS OF AUSTRALIA ANTIGEN

When Au-positive serum is ultra-centrifuged and the sediment examined by electron microscopy, regular spherules approximately 20 nm in diameter are seen. The relationship of these particles to Au is indicated by their absence in Au-negative sera and their agglutination when anti-Au antiserum is added. Since Au has been implicated in the etiology of "viral" hepatitis, the possible viral nature of the particle was suggested. Certain observations indicate that the 20 nm spherule is not a virus; first, the number of particles present per ml of serum (estimated as high as 10^{13} (9)) is far in excess of particle counts in other viremic states; second, Au particles show a wider size range (17-25 nm) (10) than observed for known viruses; third, nucleic acid has not been detected in purified preparations of the small spherules (11).

If these Au spherules are not virions, what is their nature? The possibility that they are of host origin, released in response to hepatitis virus infection, has been considered, and evidence of cross reaction with known host antigens has been reported (12). It is considered more likely, however, that these particles represent cast off protein coat (capsid) material from a "parent virus", a phenomenon observed with certain plant viruses (13).

Other morphologic forms have been observed in Au-positive serum by electron microscopy which co-aggregate with the 20 nm spherules in the presence of anti-Au, suggesting the presence of common "Au" surface antigens. The most frequently observed variant forms are "tubules", cylindrical forms approximately 20 nm in diameter and of variable length, up to several hundred nm. When present, the tubules have always been observed to coexist with the spherules. Dane observed virus-like particles 42 nm in diameter, consisting of an outer coat 7 nm in thickness, surrounding an "inner-body" of 28 nm diameter (14). Ultracentrifugation studies indicated that the larger particles are more dense than the spherules. Dane also described a "tadpole" form whose head appears identical to the 42 nm particles and from which extends a tail of variable length and resembling the tubular forms mentioned above.

Jokelainen et al (15) observed that the core of the 42 nm particle stained with uranyl acetate as visualized by electron microscopy. Such staining is consistent with the presence of nucleic acid. The 20 nm spherules did not stain. In density gradient ultracentrifugation studies Gerin et al (11) found two peaks of Au antigen immunoreactivity. The spectrophotometric absorption pattern of material in the more dense peak (density 1.39) was consistent with the presence of nucleic acid unlike material in the density 1.22 peak in which the 20 nm Au spherule was located. Jozwiak et al (16) have recently reported the results of studies of Au fractions partially purified by passage through a Sephadex G-200 column, which would not separate 42 nm Dane particles from 20 nm spherules. Data were obtained strongly suggesting the presence of nucleic acid, specifically RNA, in fractions of Au prepared in this manner.

It is suggested that the various forms described, all bearing a common "Au" surface antigen, may be related as follows; the most likely candidate for the role of primary infectious particle (virus) is the Dane particle, which probably has a nucleic acid (RNA) core; exuberant protein coat material migrates from the surface of the Dane particle in a tail-like projection, producing the "tadpole" form; the detached tails constitute the tubular forms which, in turn, fragment to the small spherules.

METHODS FOR DETECTION OF AUSTRALIA ANTIGEN

Assay procedures for Au antigen and Anti-Au antibody are of two types - screening tests and (currently) investigational tests. Screening tests, which include the micro-Ouchterlony agar gel diffusion (AGD), Counter-immunoelectrophoresis (CIEP) and Complement Fixation (CG), procedures, are described in Table 1. At the present time the electrophoretic method is used most commonly for both donor blood screening and testing of patient sera.

More sensitive methods are available, but at present are used primarily as research techniques. These include electron microscopy (17, 18), hemagglutination-inhibition (19, 20), and radioimmunoassay (21,22,23), Since the current screening methods probably fail to detect a significant fraction of Au-positive donor blood units it is expected that modifications of the hemagglutination or immunoassay methods will come into routine blood bank use within the next few years, their adoption being further hastened by the pressures of medicolegal considerations.

Table 1

ROUTINE SCREENING TESTS FOR AUSTRALIA ANTIGEN

Method	Sensitivity (cf. to Ouchterlony)	Advantages	Disadvantages
Micro-Ouchterlony (Agar gel diffusion)	1	Simplicity; ability to determine identity of antigens in different specimens	Slow - positive results may appear as late as 3-5 days; least sensitive, however, modifications may increase sensitivity considerably (24).
Counter Immuno-electrophoresis	X8-32	Rapidity (2 hrs); increased sensitivity and simplicity	Sensitivity still insufficient to detect a fraction of positive sera sera (25).
Complement Fixation	X64 or greater	Further increases in sensitivity; Ability to detect some immune complexes	Slow - best results with overnight incubation (26); Difficult assay - -Day to day fluctuation in behavior of assay -Difficulty of interpreting anti-C^1 activity of test serum -Prozone phenomenon with high titer Au; each sample must be run at several dilutions. -Some Au/Ab Complexes apparently do not bind complement (27).

AUSTRALIA ANTIGEN AND POST-TRANSFUSION HEPATITIS

The risk of Au-associated hepatitis following blood transfusion can be related to several factors:

1. Prevalence of Au in the donor population.

As shown by the data of Table 2, the prevalence of Au-positive blood in large blood banks in temperate zones of the world ranges from 0.1 to 2.0%. The principal source of Au-positive blood is the asymptomatic chronic Au-carrier; the remaining Au-positive units are contributed by persons with transient Au-antigenemia, some (but not all) of whom subsequently develop evidence of acute hepatitis (28).

The increased hepatitis risk of blood from certain donor groups is well recognized. These include prisoners, indigent paid donors selling their blood to commercial blood banks, and ghetto residents even when drug addicts and persons with a history of jaundice are excluded.

This is exemplified in the study of Goeser et al (29) comparing Au-data (Agar-gel diffusion technique) from the blood bank of a large charity hospital (Philadelphia General Hospital, PGH) and a private hospital (Hospital of The University of Pennsylvania, HUP) in the same city. Of 2156 units tested at PGH, 36 (1.67%) were Au-positive as compared to 10 of 3771 (0.27%)at HUP. The PGH bank blood included 1401 units from prisoners, of which 28 (2.00%) were positive, 128 units from commerical blood banks (2 units positive) and 31 units from "walk-in" donors offering to sell their blood (2 units positive). Among friends and relatives donating blood for PGH patients 4 of 497 (0.81%) were Au-positive as compared with 3 of 940 (0.32%) of a similar group at HUP.

In further contrast to the PGH data, among 106,294 units of blood collected from volunteer Red Cross donors in Massachusetts, 114 (0.1%) were Au positive (30). (Note also that most of the latter group were tested by the more sensitive electrophoretic method).

2. Efficiency of Screening tests in the detection of Au-Positive blood.

With the availability of the relatively rapid and inexpensive electrophoretic screening method (CIEP) and of commercially prepared antisera, almost all blood currently transfused in this country has been subjected to Au-testing (31). With appropriate modifications, the sensitivity of the CIEP test can be increased to roughly 30 times that of the Agar gel diffusion method (25).

Table 2

PREVALENCE OF Au IN BLOOD DONOR POPULATIONS

Reference	Donor Population	Number of Units Tested	Au Screening Method*	Percent Au Positive
Gocke (28) (NYC)	Not Stated	1,726	AGD	0.5
Soulier (32) (Paris)	Not Stated	10,000)	AGD	0.36
		10,000)	CF	0.40
Okochi (8) (Tokyo)	Not Stated	10,090	AGD	1.05
Sanwalt (36) (Heidelberg, Germany)	Not Stated	2,053	AGD	0.8
Kaboth (37) (Gottingen, Germany)	Not Stated	5,150	AGD	0.46
Kliman (30) (Mass.)	Volunteer (Red Cross)	106,294	CIEP	0.1
Goeser (29) (Phila.)	P.G.H. (Charity Hosp.)	2,156	AGD	1.67
	Pt. Relatives	497	AGD	0.81
	H.U.P. (Private Hosp.)	3,771	AGD	0.27
	Pt. Relatives	940	AGD	0.32
Skinhoj (33) (Copenhagen)	Not Stated	3,474)	AGD	0.06 (2 Units)
		)	CIEP	0.06 (2 Units)
		)	CF	3.26 (84 Units)
Berthold (38) (Freiburg, Germany)	Not Stated	2,583)	AGD	0.5
		)	CIEP	0.7
		)	CF	4.7
Toyoshima (34) (Tokyo)	Not Stated	3,048	AGD	0.72
		106)	AGD	0
		)	RIA	4.71 (5 Units)
		)	CF	0.94 (1 Unit)

* AGD - Agar gel diffusion; CF - Complement Fixation; CIEP - Counterimmunoelectrophoresis; RIA - Radioimmunoassay

A critical question, yet unanswered, concerns the frequency of infectious Au-positive units with Au-titers below the sensitivity threshold of the present screening tests. Preliminary data (Table 2) suggest that, at least in certain populations, a substantial number of infectious, Au-Positive units are detected only by more sensitive assay procedures. Soulier (32) observed only a slight increase in the number of Au positive units detected by a complement fixation (CF) test, as compared to AGD. In marked contrast, however, Skinhoj (33) found 84 of 3474 units positive by CF while only 2 of these were detected by AGD and CIEP. Similar data are reported by Berthold (38). (Note: The sensitivity of CF tests for Au varies widely among different laboratories. A major factor accounting for this variation appears to be differences in the complement-fixing capacity of the Anti-Au anti-sera employed).

Using an extremely sensitive radioimmunoassay (RIA), Toyoshima found 5 Au-positive units among 106 tested, only one of which was detected by CF and none by AGD (34).

3. Relationship of Au-titer to infectious risk of transfused blood

Undetected Au-positive donor blood represents a risk only insofar as the titer of Au antigen present is sufficient to produce clinical infection or, in the genetically susceptible individual, to initiate a chronic carrier state. (While the carrier state often seems harmless for the affected individual, these persons may act as a reservoir from which Au may spread by enteric or parenteral means.)

The data of Barker and Murray (35) (Table 3) suggest that at certain very low titers (high dilutions) injected Au-positive serum may no longer cause clinical disease but may still be capable of producing an Au antigenemia indicating viral replication. (The out-come of the antigenemia was not determined). In studies done in the early 1950's, Murray (39) injected 1 ml volumes of various dilutations of an icterogenic serum pool into human volunteers. Clinical and laboratory evidence of hepatitis followed injection of dilutions as high as 1 to 10,000. Serum samples from these patients, and the original serum pool were stored frozen, and in 1969 Barker et al tested these sera for Au antigen (35). The serum pool was Au-positive by CF test only (not AGD) and at the low titer of 1:10. As shown in Table 3, Au-antigenemia was detected in 2 of 5 subjects inoculated with 1 ml of a 1:10 million dilution of Au-positive serum.

Recent studies of Berthold provide information concerning the hepatitis risk of transfusion of Au-positive blood of widely varying Au-titer (38). The absolute quantitites of Au infused were considerably greater than the maximal amounts injected in the Murray's study described above. Although the number of patients was too small for firm conclusions,

TABLE 3

Incidence of Australia Antigen in

Recipients of Au Icterogenic Plasma Pool (35)

Pool Dilution	Individuals Inoculated *	Hepatitis Cases		No Clinical Illness		Au Positive Total
		No. Tested	Au Positive	No. Tested	Au Positive	
10^{0}	37	22	20	15	5	25
10^{-3}	5	2	2	3	1	3
10^{-4}	5	1	1	4	2	3
10^{-5}	5	0	0	5	2	2
10^{-6}	5	0	0	5	3	3
10^{-7}	5	0	0	5	2	2
10^{-8}	5	0	0	5	0	0

* All inoculations were 1 ml by the subcutaneous route.

the Au-titer of transfused blood bore no clear relationship to the incidence of hepatitis or Au antigenemia. (Table 4).

The data from these studies suggest that, while Au-containing serum is capable of producing clinical hepatitis at titers below the sensitivity threshold of the most potent assay techniques, only at extremely low Au levels does the infectious risk parallel the quantity of antigen injected. In practical terms it may be concluded that blood found to be even marginally positive by the most sensitive of present techniques carries a serious risk of producing post-transfusion hepatitis.

4. Protective effect of anti-Au antibody in concomittantly transfused blood

Evidence is accumulating that Au-positive hepatitis can be ameliorated or prevented by the use of specific anti-Au antibody (40,41). Although anti-Au is rarely found by AGD or CIEP in any sera except those of multiply transfused patients, by radioimmunoassay approximately 15% of healthy blood donors are found to have anti-Au in their serum (42, 43). Most studies of post-transfusion hepatitis have shown that the risk of hepatitis in a given recipient increases with the amount of blood transfused up to about 6 units. Beyond this the frequency of hepatitis ceases to rise and indeed may decline when more than 6-10 units of blood are administered. Although it has not been specifically demonstrated that this phenomenon applies to Au-positive hepatitis, the possibility seems reasonable in view of the demonstrated protective effect of anti-Au and the frequency of this antibody in the donor population.

ROLE OF AUSTRALIA ANTIGEN IN ACUTE VIRAL HEPATITIS

Prior to the discovery of Australia antigen acute viral hepatitis was attributed to infection with either a long-incubation agent (Virus B) causing parenterally-transmitted hepatitis or "serum hepatitis" and the short-incubation virus (A) producing both the epidemic and sporadic forms of non-parenterally transmitted acute hepatitis or "infectious hepatitis". The demonstration by Krugman and associates that the long incubation agent is transmissable by oral administration of serum raised the theoretical possibility of natural enteric spread of Virus B(44).

Since 1968, the sera of large numbers of patients with various forms of acute viral hepatitis have been tested for Au with the following results:

Epidemic Hepatitis: This form of short-incubation hepatitis spreads rapidly among groups of persons, especially children, under conditions of crowding and poor sanitation, and may sometimes be traced to a "point" source of fecally contaminated food or water. Au antigen studies of several such epidemics have

TABLE 4

Relationship Between Au-titer of Donor Blood and the Incidence of Post-Transfusion Hepatitis and Au-antigenemia (38)

CF-titer	No. of Au-Pos. Units CF	AGD	Hepatitis Icteric	Anicteric	Au-antigenemia within a period of six months
1 : 2	4	-	-	-	2
1 : 4	21	-	1	-	16
1 : 8	13	-	1	1	9
1 : 16	4	-	-	1	1
1 : 32	5	1	1	-	2
1 : 64	1	1	1	-	1
1 : 128	4	4	-	-	3
1 : 256	1	1	-	-	-
1 : 512	2	2	-	-	1
Total	55	9	4 and	2 (10.9%)	35 (65%)

been reported and these have been summarized by Szmuness (48). Among 428 serum samples from 11 epidemics, only two sera were Au-positive. Thus the evidence is strong that this disease is due to a virus unrelated to Au antigen.

"Serum Hepatitis": Among patients assigned this diagnosis on the basis of a history of transfusions, use of unsterile needles or other parenteral inoculations, the frequency of Au-positive hepatitis is quite variable in different series, depending primarily on the method of Au determination. (Reviewed by Shulman (19)). When the more sensitive Au-detection methods are used and serum samples are obtained soon after the onset of jaundice almost all cases are found to be Au-positive (45, 37). The MS-2 serum pool of Krugman, which has been shown repeatedly to produce long incubation hepatitis after parenteral as well as oral administration (44) was found to contain Au, as did the sera of 19 of 20 (95%) of subjects inoculated with this material (46). (By contrast, the MS-1 pool, apparently containing the epidemic hepatitis virus (virus A) produced only Au-negative, short incubation hepatitis in recipients (46)).

Sporadic "Infectious Hepatitis": It has been assumed that non-epidemic hepatitis among patients lacking a history of parenteral injections is due to "virus A", the etiologic agent of epidemic hepatitis (47). Several reports of Au studies in this disease, however, reveal that, in contrast to epidemic hepatitis, from 10-75% of such patients are Au-positive (reviewed in reference 48). Combined data from several reports show that Au was detected in 251 of 1032 (24%) of adult patients tested. Most of these sera were tested by the AGD method; when the more sensitive CF technique was used to test a series of 163 sera, 79 (48%) were shown to be Au-positive (49, 50, 51).

Although even more sensitive methods may increase the percentage of Au-positive cases, still it is likely that almost half of cases of sporadic non-parenterally transmitted hepatitis are due to other viral agents. It remains to be determined how many of these are, in fact, due to the epidemic hepatitis virus (virus A) and how many are caused by other agents, presently known or yet to be described.

When these figures concerning the frequency of Au in various forms of acute hepatitis are considered, it is evident that the terms "infectious" and "serum" hepatitis are no longer useful. As discussed above, many cases of post-transfusion hepatitis are Au-negative, especially now that Au-positive donor blood is systematically rejected. Unlike epidemic hepatitis, many cases of sporadic "Infectious" hepatitis are associated with Au. Therefore at the present time it is more appropriate to use the terms "epidemic" hepatitis and "Au-positive, or negative - Sporadic" hepatitis.

AUSTRALIA ANTIGEN AND CHRONIC LIVER DISEASE

Chronic Hepatitis:

Chronic hepatitis includes two fairly distinct subgroups, most clearly distinguished on histological grounds (52);

Chronic Persisting Hepatitis (CPH): This condition typically is manifested as a prolonged (months to years) moderate elevation of serum transaminase levels following an episode of acute hepatitis (53, 54). Usually the patient is minimally symptomatic and eventual complete recovery is the rule, with no more than mild hepatic fibrosis as a residual. Among patients with CPH 40 of 95 (42%) of reported cases have been Au positive (Table 5). Of those patients with chronic Au-antigenemia whose evaluation included liver biopsy, 5/48 (11%) had CPH (Table 6).

Chronic Aggressive Hepatitis (CAgH): This disease encompasses two somewhat distinct syndromes. First, "classical" chronic active hepatitis, typically affecting females in the adolescent to young adult age group, presenting with chronic and progressive liver disease associated with a variety of systemic features including fever, a variety of skin lesions, pleurisy, pericarditis, arthritis and arthralgias, amenorrhea, Cushingoid changes, hyperglobulinemia, positive ANA and LE preparations, smooth muscle autoantibodies and false positive serologic tests for syphilis (55). The second form of chronic aggressive hepatitis includes a less clearly defined group of patients characterized by a greater frequency of males and older patients, and a reduced incidence of systemic signs and serologic abnormalities. This group may include those patients described by Boyer and Klatskin with the histologic lesion of "subacute hepatic necrosis" during an attack of acute hepatitis, going on to a form of chronic liver disease in some cases (56).

There is preliminary evidence suggesting that Au antigen is found more frequently and smooth muscle autoantibody less often among patients with the latter type of chronic aggressive hepatitis (57-61) although one report is contrary to this trend (62).

In most series, the distinction between these two types of CAgH is not made. In a combined group of 239 patients, 87 (38%) were Au-positive (Table 5). When patients with chronic Au antigenemia were studied including liver biopsy, 17 of 48 (35%) showed the lesions of chronic aggressive hepatitis (Table 6).

Laennec's Cirrhosis:

Au does not appear to play a contributory role in the development of alcoholic cirrhosis. In 397 cases gathered from several reports by Prince (69) Au was present in only 15 (3.8%).

Table 5

Prevalence of Au-antigenemia in Chronic Liver Diseases

Author (Ref.)	Chronic Persistent Hepatitis	Chronic Aggressive Hepatitis	Primary Biliary Cirrhosis
Reinicke (63)	0/5 *	0/15	-
Kaplan (64)	8/10	2/34	0/10
Bulkley (59)	-	7/30	-
Kaboth (37)	22/28	46/75	1/17
Krassnitzky (65)	1/7	9/15	-
Becker (54)	0/14	-	-
Wright (66)	-	6/24	0/44
Fox (67)	0/19	0/32	0/39
Alarcon-Segovia (68)	-	9/18	5/7
Muller (60)	9/12	8/28	-
Prince (69)	-	-	9/10
Total (% Positive)	40/95 (42%)	87/269 (32%)	15/127 (12%)

* Number Au-Positive/Number Tested

Table 6

Chronic Liver Disease in Patients With

Persistent Au Antigenemia*

Author (Ref.)	No. of Patients	No Disease	Chronic Persistent Hepatitis	Chronic Aggressive Hepatitis
Prince (69)	7	0	1	5
Nielsen (70)	10	0	2	8
Singleton (71)	10	0	2	2
Reinicke (72)	12	12	0	0
Lebucq (73)	9	4	0	2
Total	48	16 (33%)	5 (11%)	17 (35%)

* Only patients undergoing liver biopsy are included in the tabulations.

Table 7

INCIDENCE OF AUSTRALIA ANTIGEN IN HEPATOCELLULAR CARCINOMA

Country	Author	Au-Assay Method[a]	Number Tested	Number Positive	Percent Positive
United States	Smith (77)	AGD	12	0	0
	Prince (69)	CIEP	55	2	4
	Alpert (76)	CIEP	51	3[b]	6
	Hersh (80)	CIEP	14	3[c]	21
			132	8	6.1%
Africa					
East Africa	Smith (77)	AGD	11	0	0
Uganda	Prince (69)	CIEP	34	4	12
	Vogel (79)	CF	45	18	40
Senegal	Prince (69)	CIEP	210	88	42
			300	110	36.7%
Asia					
Hong Kong	Smith (77)	AGD	42	2	5
Singapore	Simons (78)	CIEP	114	3	2.6
			156	5	3.2%

a AGD - Agar gel diffusion; CIEP - Counter-immunoelectrophoresis; CF - Complement fixation.
b All 3 of these patients were alien black males, two from Africa and one from West Indies.
c Four others of these 14 were anti-Au positive.

Hepatoma:

The relationship of Au to hepatocellular carcinoma is uncertain. Sherlock has suggested the concept of a continuous pathologic spectrum beginning with Au-positive acute hepatitis, progressing to chronic hepatitis and cirrhosis with eventual emergence of hepatocellular carcinoma (74). This concept is supported by the observations of Okochi in Japan who studied two families having a very high percentage of Au-positive members and also a high familial incidence of chronic hepatitis, cirrhosis and hepatocellular carcinoma (75).

The prevalence of Au in persons with hepatoma shows marked geographic variation (Table 7). The overall incidence among patients studied in the U.S. is 6% (8 of 132 patients; note that 3 of these 8 were aliens (76). Au was found much more often (36.7%) among African cases. It is of interest that in Hong Kong (77) and Singapore (78), areas which, like parts of Africa, have a high incidence of hepatoma, the frequency of Au-antigenemia in hepatoma cases was as low as in this country.

Preliminary data suggest a positive correlation between the presence of Au and α-fetoprotein in patients with hepatoma (79, 80).

EPIDEMIOLOGIC CONSIDERATIONS

Krugman's studies in which Au-positive (MS-2) serum was administered by mouth to human subjects demonstrated that Au-hepatitis can be spread by the oral as well as parenteral routes (44). The fact that the majority of patients with Au-positive hepatitis have had no recognized parenteral exposures is consistent with non-parenteral transmission of the virus. It is not known, however, exactly what the infectious material is -- whether feces, urine, serum, etc. As discussed above, the amounts of Au-positive serum necessary to transmit hepatitis are extremely minute (35). Some, but not all, investigators have been able to demonstrate Au in feces (81, 82, 19) and urine (82, 83). Since certain types of particles bearing Au antigenic sites are probably not infectious, the demonstration of "Au antigen" in stool, urine or other material is not direct proof of their infectious potential and unfortunately no convenient experimental animal or in vitro culture technique is available to permit study of this question.

Maternal-Fetal Transmission of Au: Au has appeared in children of Au-positive women within a few months after birth, but the route of infection is uncertain. Large numbers of cord sera and newborn sera from infants born to Au-positive mothers have been tested (85-88) and with a single exception (89) all have been Au-negative. Several infants developing Au antigenemia shortly after birth were never breast fed, and in at least one instance breast milk of an Au-positive woman was shown to contain no Au (85).

Table 8

ACTIVE IMMUNIZATION FOR Au-POSITIVE VIRAL HEPATITIS, (96)

Inoculum	Total No. Inoculated	Positive Au	Abnormal SGOT Level*	Clinical Hepatitis
MS-2 serum	25	25	24	24
Boiled MS-2 serum, 1 inoculation; Unheated MS-2 serum 4 or 8 months later	10	5 + 1†	5 + 1†	5
Boiled MS-2 serum, 2 inoculations at 4 month intervals; Unheated MS-2 serum 4 months after 2nd inoculation	4	1†	1†	0

* SGOT, serum glutamic oxaloacetic transaminase.
† Au was detected and SGOT level was abnormal on only one day.

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