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Public Health Utility of Premarital Screening Tests for ABO / Rhesus Blood Group Incompatibilities and Hepatitis B and C Viral Infections

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Authors' contributions

This work was carried out in collaboration between both authors. Both authors read and approved the final manuscript.

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ABSTRACT

Background: Premarital medical screening tests (PMST) occupies a strategic public health position in the prevention and control of both heritable and infectious health conditions which a couple who intend to get married may transmit to each other and also pass to their offspring. It is useful in the detection of such infectious diseases like hepatitis B virus and hepatitis C virus; as well as genetically inherited diseases like haemolytic disease of the new born (HDN) resulting from ABO and rhesus blood group incompatibilities. This study was aimed at determining the public health roles of PMST, by ascertaining the prevalence of viral hepatitis A and B viral infections and ABO / rhesus incompatibilities in premarital couples.
Methodology: This is a retrospective assessment of laboratory records of premarital screenings for 252 males and 252 females who were preparing to get married. The panel of tests include ABO and

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Rhesus (D) blood groups, hepatitis B surface antigen and hepatitis C. The procedure for all the analysis, followed the directives contained in the laboratory standard operating manual.
Results: The overall prevalence of the viral hepatitis infections (B or C) was found to be 1.8% among the 504 persons whose records were reviewed. The highest prevalence of 3.2% among age brackets was noticed among the >35-year-olds, while the least prevalence 0f 0.6% occurred among the 31–35-year-olds. The females had a higher prevalence (2.4%) than the males (1.2%). The prevalence of HBV among the premarital individuals was 1.4%; while hepatitis C was 0.4%. The prevalence of incompatibilities for ABO and rhesus factor were 33.3% and 5.6% respectively.
Conclusion: The public health utility of PMST is obvious, as widespread screening by prospective couples will significantly reduce the incidences of many genetic disorders, sexually transmitted, blood-borne and vertically transmitted infections. The use of legal enactments and massive public enlightenments are recommended to ensure widespread compliance to premarital screening by prospective couples.

Keywords: ABO bold groups, Hepatitis B virus; Hepatitis C virus; premarital medical screening; rhesus incompatibility.

1. INTRODUCTION

Premarital medical screening tests (PMST) are panels of laboratory analysis conducted on couples as part of preparations for living together as husband and wife. They are useful in identifying heritable and infectious disease conditions which a couple may pass to their offspring. PMST gained prominence in Nigeria in the early 1990s following apparent high incidence HIV among newly married couples which threated family cohesions. Today PMST occupies a strategic position in the prevention of such infectious diseases as hepatitis B virus and hepatitis C virus; as well as genetically inherited diseases like haemolytic disease of the new born (HDN) resulting from ABO and rhesus incompatibility [1].

The ABO and the Rhesus (Rh) blood group systems discovered by Karl Landsteiner in early 1900s remain the most clinically significant blood group antigens among several blood group antigens located on the red cell membrane [2,3]. The importance of blood group investigations as part of the PMS is hinged on the need to prevent Rh incompatibility which is commonly associated with neonatal jaundice, *erythroblastosis fetalis* or haemolytic diseases of the neonate (HDN), hydrops fetalis, and stillbirth [2]. The ABO incompatibility which is more prevalent than rhesus incompatibility is not much of a public health threat because ABO incompatibility results in much milder conditions which, unlike rhesus incompatibility does not require prophylactic treatment [4,5].

Rhesus (Rh) incompatibility is commonly found in unions of Rh-negative women and Rh-positive men who had Rh positive babies. The foetus with Rh D antigen on the cell membrane is able sensitize the Rh-negative mother whose red blood cells have no Rh D antigens to produce anti-D antibodies against the Rh antigens of the foetus, this is alloimmunization. The antigens may then bind to the Rh-positive red cells and have them destroyed. This usually arises due to transplacental or foeto-maternal hemorrhage which may occur during pregnancy or childbirth. Destruction of the babies' red cells by maternal immunoglobulin (IgG) antibodies causes the alloimmune hemolytic disease of the neonate, which may not be severe in the initial sensitization, but in subsequent pregnancies there are severe complications, such as intrauterine hydrops fetalis [4,5]. Incompatibility in ABO blood groups, mostly found among offspring of blood group O mothers and non-blood group O fathers, though, in rare cases, has been reported in unions between groups A and B parents [5].

Hepatitis B and hepatitis C viral infections pose great global public health threats as a result of their chronicity, high morbidity and mortality resulting from cirrhosis and hepatocellular carcinoma and other associated conditions [6]. With an estimated 1.1 million deaths in 2022 arising majorly from complications of chronic hepatitis B virus (HBV) infection, like cirrhosis and hepatocellular cancer, HBV infection is a leading cause of death worldwide. The World Health Organization projected number of people living with chronic hepatitis B in 2022 is 254 million [7,8].

The World Health Organization estimated number of persons living with chronic hepatitis C by the year 2023 is about 50 million people globally, with an approximated 1 million new

infections yearly [9,10]; and about 3.26 million children affected by chronic hepatitis C infection. highlighting its significance as a burden [10,11,12]. HCV is essentially a blood-borne pathogen transmitted horizontally through percutaneous exposure to infected blood, with lesser transmission vertically by mother to child, or sexual intercourse, especially among men having sex with men [10]. The pathogen is not known to penetrate an intact skin, but can be transmitted where infected blood makes contact with mucosal surface such as the eyes [10].

The commonest modes of HBV transmission are vertically from mother to child, sexually via vaginal, oral, anal or other forms of sexual contact, and parenterally through contact with blood or blood products. The virus highly stable at 37 °C on environmental surfaces for more than 22 days, and is detectable in blood and body fluids including saliva, tears, sweat, semen, and vaginal secretions of infected individuals. In areas with low prevalence of hepatitis B infection, transmission is mainly through injectable drug uses and high-risk sexual behaviours [13].

Pre-marital medical laboratory investigations have potentials for lots of positive impacts and opportunities for positive public health outcomes. Early detection of infectious diseases such as HIV, syphilis, and hepatitis; and the determination of incompatibilities that may result in genetic disorders, such as haemoglobinopathies and HDN will enable timely and informed decisions on preventive measures. It will lead to reduction in the incidence of such infectious and non-infectious conditions [14]. Premarital testing goes together with premarital counselling, thus enhances public health knowledge and awareness among persons intending to start new families. It enables he understanding of possible risks of children inheriting genetic conditions, engenders open communication and encourage mutual support in the handling of health issues [14].

There were inadequacy of studies on the prevalence of ABO and Rhesus Blood Group Incompatibilities and Hepatitis B and C Viral Infections among prospective couples in Port Harcourt.

This study was therefore aimed at reviewing premarital screening tests of ABO / rhesus blood groups, hepatitis A and hepatitis B viral infections, determine the prevalence of incompatibilities and seropositivities as applicable and make relevant recommendations for the enhancement of public health.

2. MATERIALS AND METHODS

2.1 Study Design, Setting and Study Subjects

This is a retrospective analysis of laboratory records of premarital screenings for couples who were preparing to get married. The panel of tests include ABO and Rhesus blood group, hepatitis B surface antigen (HBsAg) and anti-hepatitis C (HBV) tests, which were conducted at Diagnostix and Scientifique Laboratories Port Harcourt, which renders services to public and private healthcare facilities, between January 2020 and December 2023.

Most of the couples were referred for the screenings, by religious bodies or families who are increasingly being aware of the traumatic experiences of parties and families over avoidable genetic conditions or infectious diseases, as a result of marriages contracted in ignorance of life-threatening conditions that may be prevented by going for screenings. Others were couples and individuals who wanted to verify their statuses before informing their families or places of worships.

2.2 Laboratory Analysis for HBsAg and Anti-HCV

The procedure for HBsAg and anti-HCV screening, was in accordance to the directives contained in the laboratory standard operating manual. Three to four mililitres of blood were collected from the subjects and allowed to stand in test tubes until clotted, before being centrifuged at 3000 revolutions per minute for five minutes. The sera were tested using one-step HBsAg test strip (SD Bioline) and a one-step HCV test strip (SD Bioline), respectively, following the manufacturer instructions. The sensitivity and specificity of rapid test kits of HBsAg and one-step HCV test strips were 99.1% and 99.6%, respectively.

2.3 ABO Blood Grouping and Rhesus (RH) Typing

The procedure for ABO and Rh blood groups screening, also followed the recommendations contained in the laboratory standard operating manual. Three to four millilitres of blood were collected from the subjects and transferred to

EDTA containers. ABO and Rh blood groups determinations were carried out with the whole blood samples on a white slide using monoclonal blood grouping antisera; anti-A, anti-B, anti-AB, and anti-D (BIOTEC Laboratories Ltd, Great Britain), in compliance with the manufacturer's instructions.

2.4 Statistical Analysis

Data were entered and clarified using Microsoft Excel version 16, then analyzed using IBM SPSS Statistics version 25. Associations between infections with HBV, HCV and HBV/HCV on one hand and age, gender, and blood group indices on the other hand were tested for significance using Chi square test of independence at 0.5 level of significance.

3. RESULTS

A total of 504 medical laboratory records belonging to 252 premarital couples were reviewed and relevant data on results of ABO and Rhesus blood groups, HBV and HCV were extracted and analyzed. The ages of the subjects ranged from 19 years to 46, with a mean of 30.85 ± 5.452 years, the median is 31.00 and the mode is 34 years. The ages of the 252 females were from 19 to 41 ± 4.623 years, the mean age is 28.05 ± 0.993 years, the mode is 26 years and median 27.00 years; the ages of the 252 males ranged from 24 to 46 years, the mean was 33.65 ± 4.737, the median age was 34.00 ± 5.007 years and the modal age was 34 years (Fig. 1).

3.1 Distribution of ABO and Rhesus Blood Groups in Premarital Screening Tests

The blood group O Rh (D) +ve was the dominant of all the blood groups amounting to 59.7% of all blood groups, followed by A Rh (D) +ve (17.5%), B Rh (D) +ve (15.1%), AB Rh (D) +ve (1.6%), there was no AB Rh (D) -ve observed in the analysis. There were 93,8% RH D positive persons and 6.2% RH D negative. Rhesus (D) incompatibility, denoted as a union between a RH (D) Positive Males and RH (D) Negative Females, was observed in 5.6%, while 94.4% were found to be compatible. ABO incompatibility was 33.3% while 66.7% were compatible (Table 1).

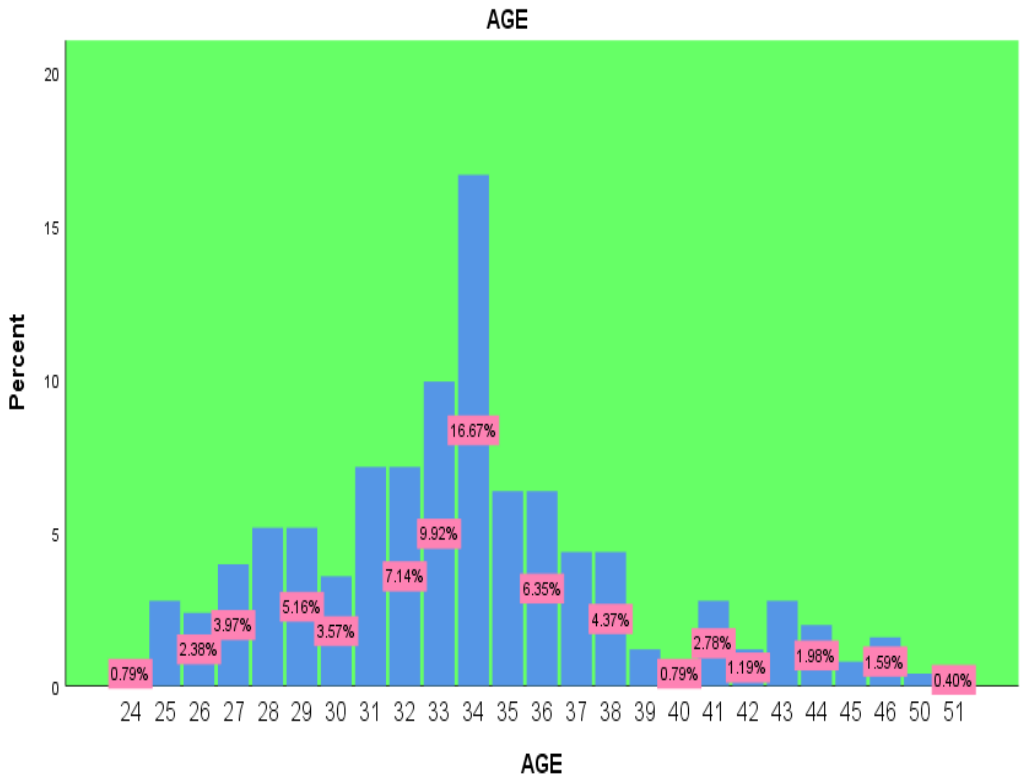

Fig. 1. Frequency and distribution of the ages of subjects

Table 1. Frequency and distribution of blood groups of premarital couples

Characteristics	Prevalence	Percent
ABO / RH Groupings		
O Rh (D) +ve	301	59.7
A Rh (D) +ve	88	17.5
B Rh (D) +ve	76	15.1
AB Rh (D) +ve	8	1.6
O Rh (D) -ve	20	4.0
A Rh (D) -ve	7	1.4
B Rh (D) -ve	4	0.8
ABO Groupings		
O	325	64.5
A	96	19.0
B	14.9	14.9
AB	8	1.6
RHESUS Groupings		
Positive	473	93.8
Negative	31	6.2
Total	**504**	**100.0**
ABO Unions		
O/O	108	42.9
O/A	60	23.8
O/B	48	19.0
O/AB	6	2.4
A/A	7	2.8
A/B	16	6.3
A/AB	2	O.8
B/B	5	2.0
ABO Compatibilities		
Compatible	168	66.7
Incompatible	84	33.3
Rhesus (D) Unions		
Rh(D) Positive/ Rh(D) Positive	222	88.1
Rh(D) Positive Males / Rh(D) Negative Females	14	5.6
Rh(D) Positive/ Females / Rh(D) Negative Males	15	6.0
Rh(D) Negative / Rh(D) Negative	1	0.4
Rhesus Compatibilities		
Compatible	238	94.4
Incompatible	14	5.6
Total	**252**	**100.0**

3.2 Prevalence of HBV and HCV among Premarital Couples

The overall prevalence of the viral hepatitis infections (B or C) was found to be 1.8% among the 504 persons whose records were reviewed. The highest prevalence of 3.2% among age brackets was noticed among the >35-year-olds, while the least prevalence 0f 0.6% occurred among the 31–35-year-olds. The females had a higher prevalence (2.4%) than the males (1.2%). The highest prevalence among the ABO/RH blood groups occurred among the B RH D positive group (2.6%); and within the ABO group, B recorded the highest prevalence of 2.7%. Within the Rhesus group, the RH (D) positive had 1.9% prevalence and zero for RH (D) negative individuals (Table 2).

3.3 Prevalence of HBV among Premarital Couples

The prevalence of HBV among the premarital individuals was 1.4%; the highest prevalence of 3.2% within the age brackets was found among >35 years bracket the 18–25-year-olds recorded zero prevalence. The females had higher infection rates of 1.6% than the males. Blood groups B (2.6%) and O (1.2%) were the only ones that had positive cases among ABO, while RH D positive had prevalence of 1.5% and RH D negative had zero prevalence (Table 3).

Table 2. Frequency and prevalence of HBV and HCV in premarital screening tests

Characteristics	Number Tested	Positive	Prevalence %
Age Brackets			
18-25	91	2	2.2
26-30	153	3	2.0
31-35	166	1	0.6
>35	94	3	3.2
Gender			
Males	252	3	1.2
Females	252	6	2.4
ABO RH Blood Groups			
O RH D Positive	301	6	2.0
A RH D Positive	88	1	1.1
B RH D Positive	76	2	2.6
AB RH D Positive	8	0	0.0
O RH D Negative	20	0	0.0
A RH D Negative	7	0	0.0
B RH D Negative	4	0	0.0
ABO Blood Groups			
O	325	6	1.8
A	96	1	1.0
B	75	2	2.7
AB	8	0	0.0
Rhesus (D) Blood Groups			
RH (D) Positive	473	9	1.9
RH (D) Negative	31	0	0.0
Total	**504**	**9**	**1.8**

Table 3. Frequency and prevalence of HBV in premarital screening tests

Characteristics	Number Tested	Positive	Prevalence %
Age Brackets			
18-25	91	0	0.0
26-30	153	3	2.0
31-35	166	1	0.6
>35	94	3	3.2
Gender			
Males	252	3	1.2
Females	252	4	1.6
ABO RH Blood Groups			
O RH D Positive	301	5	1.2
A RH D Positive	88	0	0.0
B RH D Positive	76	2	2.6
AB RH D Positive	8	0	0.0
O RH D Negative	20	0	0.0
A RH D Negative	7	0	0.0
B RH D Negative	4	0	0.0
ABO Blood Groups			
O	325	5	1.5
A	96	0	0.0
B	75	2	2.7
AB	8	0	0.0
Rhesus (D) Blood Groups			
RH (D) Positive	473	7	1.5
RH (D) Negative	31	0	0.0
Total	**504**	7	1.4

Table 4. Frequency and prevalence of HCV in premarital screening tests

Characteristics	Number Tested	Positive	Prevalence %
Age Brackets			
18-25	91	2	2.2
26-30	153	0	0.0
31-35	166	0	0.0
>35	94	0	0.0
Gender			
Males	252	0	0.0
Females	252	2	0.8
ABO/RH Blood Groups			
O Rh (D)+ve	301	1	0.3
A Rh (D)+ve	88	0	0.0
B Rh (D)+ve	76	1	1.3
AB Rh (D)+ve	8	0	0.0
O Rh (D)-ve	20	0	0.0
A Rh (D)-ve	7	0	0.0
B Rh (D)-ve	4	0	0.0
ABO Blood Groups			
O	325	1	0.3
A	96	0	0.0
B	75	1	1.3
AB	8	0	0.0
Rhesus (D) Blood Groups			
RH (D) Positive	473	2	0.4
RH (D) Negative	31	0	0.0
Total	**504**	**2**	**0.4**

3.4 Prevalence of HCV among Premarital Couples

A prevalence of 0.4% was found for HCV among the 504 premarital individuals; a group prevalence of 2.2% was observed within the18–25-year-olds bracket, while all the other age brackets recorded zero prevalence. The females had a prevalence rate of 0.8%, but the males had a zero prevalence. Blood groups B (1.3%) and O (0.3%) were the only ones that had positive cases among ABO; RH D positive had prevalence of 0.4% and RH D negative had zero prevalence (Table 4).

4. DISCUSSION

The utility of early detection of diseases provided by premarital screening of prospective couples provides enormous prospects to prevent the passage of diseases among couples and to their unborn children. It has the public health benefit of reducing the incidence of infectious diseases such as viral hepatitis and genetic conditions associated with rhesus incompatibility. This study has been able to establish the prevalence of HBV and HCV infections, as well as ABO and rhesus incompatibilities among premarital couples.

The prevalence of Rhesus negative blood group in this study was 6.6%; this is higher than the prevalence of 3.0% obtained in a similar study in nearby Yenegoa [15]. It however aligned closely with the outcomes 5.3% reported in a related studies in Ekiti state [16], the 6.2% observed among reproductive age women in Ethiopia [17] and the prevalence of 6.4% [2] in another Ethiopian study [2]. The union between Rh (D) positive males and Rh (D) negative females are considered as incompatible because of the chances of alloimmune haemolytic disease of the fetus and newborns of the union, resulting in the destruction of the red blood cell of the neonates by maternal immunoglobulin (IgG) antibodies that crosses into foetal circulation within the womb. A more serious form of the condition occurs when the maternal alloantibodies are directed against the D antigen of the Rh blood group system due to the high immunogenicity of D antigen [3]. While the risk of alloimmunization can be successfully managed by the use of Rh D prophylaxis, in some parts of the world, the problem remains intractable in sub-Saharan Africa due to a number of factors such as deficient or non-existence of good quality medical facilities, insufficient manpower, scarcity or unaffordability of anti- Rh D immunoglobulin, multidimensional poverty, ignorance and superstitious beliefs.

The prevalence rates of 1.8%, 1.4% and 0.4% recorded in this study for the overall (HCV or HCV), HBV and HCV respectively were low compared with results obtained elsewhere in the general population and designated populations, such as the prevalence rates of 10.4%, 7.0% and 4.0%, respectively, for the corresponding parameters in an Ethiopian study among prison inmates [18], the prevalence of 10.6% and 2.7% for HBV and HCV apiece in an Iranian study [19], but much higher than the overall prevalence of 0.6%, with HBV 0.52% and HCV 0.05% in a premarital screening study in Saudi Arabia [1]. Several studies across countries have shown disparate results like a Tanzanian study that reported 3.5% apiece for HBV and HCV, with an overall prevalence of 6.3% [20]. A study in Port Harcourt reported a prevalence of 5.8% for HBV and 0.5% for HCV [21], while another reported 6.0% for HBV and 0.7% for HCV among blood donors [22]. While the prevalence of viral hepatitis appears to be reducing in some areas, the pathogens are very much around and contributing to morbidity and mortality.

In this study, the seroprevalence of HBV and HCV were found to be highest in the above 35 years bracket, which was consistent with an Ethiopian study [23]. This however differs from the results of another Ethiopian study where the highest prevalence rates were found among the 21–30-year-olds [24]. This study differed from a number of previous studies where males were found to have higher prevalences than females [1,22,23,25], the results however was in alignment with a study carried out in Ethiopia [24].

The blood group B recorded more prevalence rates of all HBV and HCV infections than other ABO groups and followed by the blood group O. This aligned partly with a Nigerian study where persons of B blood group were found to have an insignificantly ($P > 0.05$) higher prevalence of HBV infection [26]. One the other hand, a study in Pakistan reported a stronger association between HCV and blood group O than other blood groups [27]. We found the associations between blood groups and the viral infections to be statistically insignificant ($P > 0.05$), more in-depth studies are required in this regard. While studies in Nigeria and elsewhere have reported strong association between Rhesus factor negative individuals and viral hepatitis [26,27]; none of the Rh (D) negative persons in this study were found be infected, though there was no statistical association ($P > 0.05$). The reason may be due to the small number of rhesus negative persons compared to the rhesus positive.

The prevalence rates of HDN due to ABO blood group incompatibility in this study is 33.3%, which is higher than the 5.6% for rhesus; this corroborates data established in previous studies on the higher prevalence of ABO related HDN [28-31]. Rhesus incompatibility has predominantly received more attention than ABO incompatibility, due the high morbidity, mortality costs in human and material resources associated with the HDN and other outcomes of rhesus incompatibility. It is important to state that haemolytic disease of newborn is also associated with ABO groups.

ABO-related HDN results from transplacental passage of immunoglobulin (Ig)-G antibodies of the mother specific for the blood group in the foetal circulation. The outcome is haemolysis of the foetal red blood cells, with consequent anemia and hyperbilirubinemia of the foetus. The union of A or B father with O blood group mother with immune anti-A and anti-B antibodies are reported to be most likely cause of ABO associated HDN, though the pathogenesis remains unclear [28-31]. It is not usual for Rh incompatibility to affect first pregnancies when maternal sensitization occur, subsequent pregnancies are mostly affected with attendant severe complications. ABO incompatibility, on the other hand affects first pregnancies as a result of preexisting maternal antibodies [5].

Overall, this study did not find significant statistical relations between the socio-demographic characteristics and infections of hepatitis B and C among premarital couples in port Harcourt, Nigeria. While the results were consistent with the outcomes of some of the results obtained elsewhere, there were inconsistencies with some other outcomes. The inconsistencies observed in the outcomes of this study may be ascribed to several factors relating to study design, sample population, test methods, sample size, variations in geographical position, cultural, behavioural and socioeconomic situations and so on.

The study had the limitation of relying on secondary data which was expectedly associated with incompleteness, as only information required for clinical use was available. There may also be inadequate record keeping, and the test methods were limited to what was required

in a clinical diagnosis as contrasted with original study design.

5. CONCLUSION

The public health utility of premarital testing is not in doubt. Many countries particularly in Middle East where endogamous and consanguineous marriages are common have laws enacted to make premarital screening programmes mandatory for couples before obtaining marriage certificates. This is as a result of high prevalence of autosomal recessive genetic diseases such as cystic fibrosis, sickle cell disease, and thalassemia, as well as sexually transmitted and blood borne infections. Widespread acceptance of PMST will go a long way in reducing the incidence of infectious diseases like viral hepatitis, and conditions like HDN which is a fallout of rhesus incompatibility. It is advocated that this can be done through legislative enactments and public health enlightenment campaigns to ensure that a critical mass of the population embraces premarital screening prior to commencement of the marital union. This will lead the public health benefits of reducing the prevalence of these diseases; and engender a healthier population. The use of legal enactments and massive public enlightenments are recommended to ensure widespread compliance to premarital screening by prospective couples.

DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of this manuscript.

CONSENT

It is not applicable.

ETHICAL APPROVAL

Written approval was obtained from the management of Diagnostix and Scientifique Laboratories, Port Harcourt. The Study was conducted in accordance with the Helsinki declaration.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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