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Nucleic Acid Testing: An Adjunct to Screening Modalities in Blood Banking: A Study in a Govt. Medical College

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Abstract: The purpose of any blood/ blood product, when needs to be transfused to any patient, must be a safe one. For that reason, mandatory screening for diseases like HIV, HBV, HCV, Malaria & syphilis are done in all the authentic blood banks in Odisha. Conventionally screening tests for these diseases are usually done by rapid tests/ ELISA tests. During last few years to lessen the window period of these diseases, more advanced test modality like NAT (Nucleic Acid Testing) has been introduced in some transfusion centers in India. The present study has been done to find out the benefit & limitation of the NAT test over ELISA test. The study has been carried out over a period of two years in VIMSAR, NAT center of Odisha where all ELISA negative samples are subjected for testing. Some seronegative cases in ELISA when subjected to NAT testing showed positive results. The percentage of positive samples in NAT which have showed negative in ELISA test was found out to be 0.08% in HI, 0.27% in HBV &0.01% in HCV. NAT Screening may be beneficial in the population where sero-prevalence of Transfusion transmitted infection is high. Although NAT has got an added advantage of reducing the window period, all the blood centers of India cannot afford for the cost of NAT test, so proper donor deferral should be adopted during selection of eligible donors.
Keywords: NUCLEIC ACID TESTING, Window Period, Blood Safety.

INTRODUCTION

Nucleic Acid Testing is a molecular technique introduced in developed countries for screening transfusion transmitted infection as early as in late 1990s. The principle is based on amplification of targeted regions of viral RNA or DNA & thereby detecting them.

NAT is a highly sensitive & sophisticated screening modality which has reduced the window period of HBV to the extent of 10.34 days, HCV to 1.34 days & HIV to 2.93 days in comparison to serological tests.

During last one decade most of the developed countries have introduced NAT testing for screening HIV, HBV, & HCV infections. It is claimed to be a highly specific & sensitive test. It also curtails the window period for these diseases to a significant degree. But since it is a costly test for screening, in India most of the blood banks still depend on Rapid test / ELISA test. There are two types of NAT testing namely IDNAT & MPNAT.

In India till now Blood transfusion service has not been streamlined. In outdoor voluntary blood donation camps due to huge crowed pulling, proper donor screening is not possible. Moreover though it is illegal to entertain professional donors, they still continue-to donate blood.

In most of the blood banks in Odisha the TTI testings are neither quality assured nor well validated. Due to fund deficit most of the blood banks cannot meet the expenses for calibration of the major equipments. In Odisha earmarked trained dedicated blood bank officers are not posted in the Blood Banks. Even for transfusion & preservation of blood/ blood products, cold chain is not so effective due to various reasons like mishandling, power cut etc. Moreover in all most all hospitals, there is unnecessary over use of blood/ blood products.

MATERIALS & METHODS

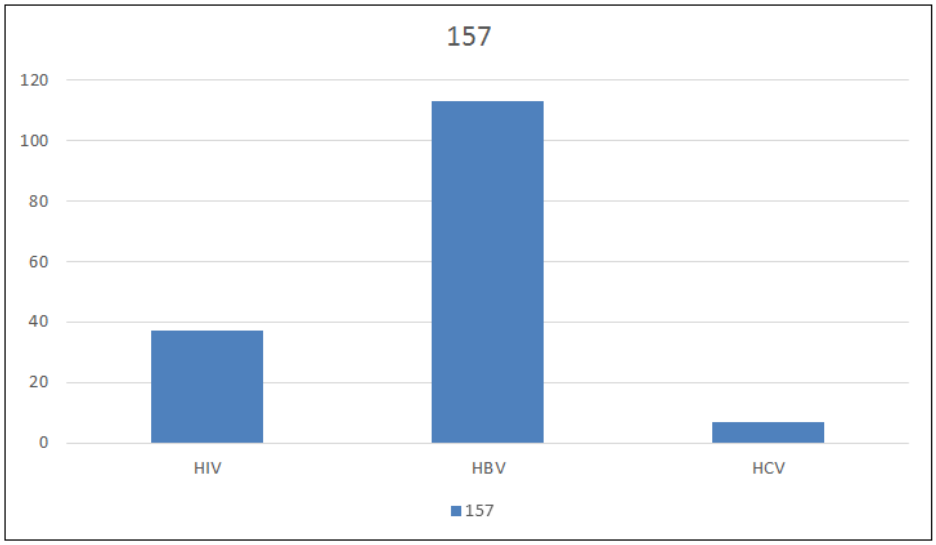
Facility for Nucleic Acid Testing by PCR technique is available in 3 old Govt. Medical Colleges namely SCB Medical College, Cuttack, VSSIMSAR, Burla & MKCG Medical College, Berhampur and capital hospital, Bhubaneswar. Another two blood banks namely B.M.C hospital BBSR and Central Red Cross Blood Bank, Cuttack are attached with these NAT centers. In our Medical College, NAT Laboratory was established in June 2016.

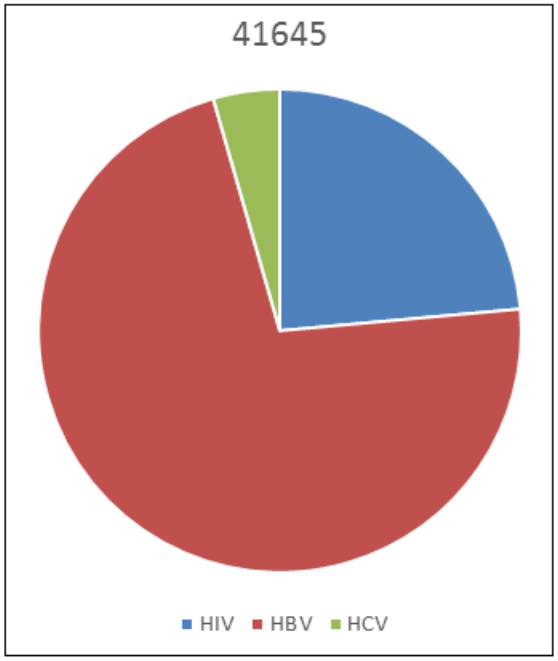
All the samples screened for HIV, HBV & HCV by Nat machine in our center over a period from June 2016 to October 2018 were taken into consideration. As a protocol all the donors' samples collected in house / in VBD camps were first subjected to ELISA test & all ELISA negative samples were subjected for NAT screening. All sero-positive samples detected in ELISA were discarded.

Before collection of blood from the donors, proper physical examination and parameters of the donor including wt, age ,Hb% were done and other relevant questionnaires as per NACO guideline were asked to declare a donor as fit to donate. All these eligible donors were counselled to be repeat donors. Display of self-deferral was done at camp site. Consent of donor before collection of blood was taken in each case.

RESULTS

Total samples tested from June 2016 to October 2018 by Nat technique is 41,645. Out of these HIV positive is 37, HBV positive is 113, HCV positive is 7.





DISCUSSION

In India family/ replacement donors still provide more than 45% of the collected blood. But due to increased awareness for voluntary blood donation proper screening of TTIs testing should be done.

Some centers use enhanced chemiluminisence Immunoassay (EC) for detection of HBsAg, anti HIV & anti HCV in donor's serum.

Family donors are not safe as voluntary donors & voluntary non remunerated repeat blood donors are safer than the first time blood donors [1, 6].

In a Malaysian study of 1388 donor samples which were screened for both serology & NAT, 1.37% samples showed sero-positivity in ELISA, but were non reactive by NAT. On confirmatory tests, these samples were found to be false reactive [2].

In Germany for screening HCV, MP NAT screening was first introduced. No doubt it is cost effective in comparison to ID-NAT, but it has its own disadvantages also. The whole size of pooled blood detection is blocked until the NAT report is available. Also as viral nucleic acid concentration gets diluted in large pool of samples the sensitivity of NAT may decrease & if a pool is tested reactive, the whole blood requires resolution to identify the single positive unit & this process requires an additional step of handling, additional time for testing & hence delay in the release of units. Indivisual NAT screening seems to be more sensitive. But on the other hand it is not economical [3]. In United Kingdom, NAT has reduced the risk of HCV by 95% & the HIV by 10%.

The introduction of NAT for HBV screening & HBV vaccination policy has reduced the risk of HBV infection [4].

In a developing country like India introduction of NAT in blood bank screening is a debatable decision. Because for NAT screening of each blood unit the cost is approximately Rs750/- whereas the cost for an ELISA screening is about Rs 200/-. In addition to it for establishment of a NAT Laboratory, infrastructure cost, manpower cost & cost for manpower training is also required. In NAT PCR testing the window period for HIV, HBV & HCV is shortened by 8 days, 9 days & 6 days.

In a study conducted at ICRC blood center, Mumbai, seroprevalence in blood donors was found to be for HIV 0.08%, for HBV 0.7% & for HCV 0.09% [5]. In a study by Markoo *et al*., 8 samples out of 12224 seronegative samples were NAT positive.

In a study in NORTH India ID NAT results were compared with serological methods for 37,898 samples. Out of these 1.49% were reactive by NAT, HIV-1 (0.09%), HCV (>25%) & HBV (0.08%).

In a study by Stramer *et al*., false positively of NAT has been reported to be about 1 in 15, 800 units for HIV & HCV NAT. this false positivity may be due to cross contamination [10].

Nucleic acid technique is a highly sensitive & advanced technique which has reduced the window period of HBV, HIV, HCV but it involves high cost, dedicated infrastructure facility, equipments, consumables & technical expertise.

In HIV-1 testing by NAT, some give false positive results due to combined heterogeneity of HIV virus or due to low viral load.

Since ELISA does not pick up seropositivity in HBV infection in occult hepatitis, non sero converting or delayed sero converting individuals, NAT-PCR test is the best option in these cases. Increased heterogeneity of HIV may sometimes give false negative results. There is incidence of transfusion of HIV-1 infection by NAT negative samples with low viral load.

Nucleic Acid technique also gives false positive result due to cross-contamination. Nucleic Acid test is essential- for Acute Hepatitis. External quality control for NAT PCR is not available.

Since India is a under-resourced country, regional blood canters may be established at specific blood bank. Poor & defective method of transportation of blood samples is also a real concern for NAT test. Therefore quality management & good manufacturing practice are very important to exclude false positive cases.

No single test is full proof

NAT PCR test has its own limitations. It only detects the presence of viral RNA/DNA. At times the viral load is also low to be detected by ELISA. So NATPCR can be an adjunct to ELISA, not a substitute for ELISA test.

The chance of transmission of seropositive samples for HIV, HBV & HCV in India is high in multi transfused thalasemia patients due to window period transmission [11]. It is still a debatable question as to which NAT, MP NAT/ ID NAT is better.

In NAT, PCR also there is risk of transmission of window period infection. But it definitely reduces window period for HIV, HBV & HCV [12].

In a developing country like India, stringent donor screening is preferable to Nat PCR installation in all the blood banks.

The need for NAT depends on prevalence & incidence rate of infection in blood donor population, available resources and infrastructure.

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