Appendix B

FDA Systematic Literature Review: HPA and HNA Devices

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1. Methods

FDA conducted a systematic literature review to assess the safety and effectiveness of FDA-cleared HPA and HNA devices. Safety and effectiveness of these devices is related to the accuracy of the test results used in the transfusion setting and for disease diagnosis. Every effort was made to include articles examining the analytical performance of FDA-cleared HPA and HNA devices in this review, as analytical performance determines safety and effectiveness.

In this literature review, we included articles that used data generated from FDA-cleared HPA and HNA devices.

We sought to address the following questions through systematic literature review:

- What is the reported analytical performance of the HPA devices (HPA typing devices and HPA antibody detection devices)?
- What is the reported analytical performance of the HNA devices (HNA typing devices and HNA antibody detection devices)?
- How are the HPA and HNA testing results used to aid blood transfusion?

1.1. Literature search for the analytical performance of FDA-cleared HPA devices and their use to aid in transfusion

We searched electronic databases PubMed and EMBASE using the following terms for all available articles published before May 01, 2017:

- ("Antigens, Human Platelet") OR ("Antibodies, Human Platelet") OR "human platelet antigens" OR "platelet specific antigens" OR "platelet alloantigens" OR "HPA" OR "human platelet antibodies" OR "human platelet alloantibodies" AND (test OR tests OR testing OR assay OR detect OR determination OR kit OR kits OR "PIFT" OR immunofluorescence OR "monoclonal antibody immobilization of platelet antigens" OR "MAIPA" OR "solid phase red cell adherence" OR "SPRCA" OR "monoclonal antigen capture" OR "Fluorescent Antibody Technique, Direct" OR "Enzyme-Linked Immunosorbent Assay" OR platelet matching) AND ("Blood Transfusion") AND ("Respiratory Distress Syndrome, Adult" OR adverse OR risk OR risks OR complication OR safety OR "side effect" OR "TRALI" OR "transfusion related acute lung injury" OR "ARDS" OR "acute respiratory distress" OR survival OR false negative OR false positive OR clinical outcome OR error OR incorrect OR mistyped OR invalid OR defect OR discordant OR accuracy OR success OR efficacy OR effective OR efficiency OR platelet transfusion refractoriness OR thrombocytopenia OR neonatal alloimmune thrombocytopenia OR immune thrombocytopenia OR post-transfusion purpura)

The search terms include the type of tests used to detect human platelet antigens and human platelet antibodies, as well as the diseases clinically impacted by HPA testing results such as transfusion-related acute lung injury (TRALI), platelet transfusion refractoriness (PTR), immune thrombocytopenias, fetal/neonatal alloimmune thrombocytopenia (FNAITP) and post-transfusion purpura (PTP).

We limited our literature search to full text articles published in English from January 1, 1994 to May 1, 2017. We believed that a review of the articles published in the last 23 years would cover the information needed to evaluate the analytical performance, safety and effectiveness of HPA devices. Conference abstracts were excluded from the literature review.

The search generated 343 publications. Titles and abstracts were reviewed to exclude studies not related to safety, effectiveness or performance of FDA-cleared HPA devices. Studies conducted in the US or in other countries were included if they used FDA-cleared HPA devices. However, the literature does not typically use the term "FDA-cleared" when referring to the HPA assays used in studies. We compared the HPA assay name, type, and manufacturer information provided in each article with our database of cleared HPA devices to determine whether a study used an FDA-cleared assay(s).

Many studies referencing HPA typing were excluded as they did not specifically indicate the use of an FDA-cleared device (Figure 1).

A total of 312 articles were excluded during the initial screening for the following reasons:

- Animal study
- Not specific to FDA-cleared HPA devices
- Not in English Language
- Inadequate analytical performance or clinical use information

After titles and abstracts were reviewed, the full-texts of the remaining 31 articles were further examined for eligibility. A total of 16 articles were excluded during the full text review using the same criteria described above.

A total of 15 articles were included for systematic review. Additional information regarding the methodology for inclusion and exclusion criteria may be found in Figure 1.

1.2. Literature search for the analytical performance of FDA-cleared HNA devices

We searched electronic databases PubMed and EMBASE using the following terms for all available articles published before May 01, 2017:

- ("Antigens, Human Neutrophils") OR ("Antibodies, human neutrophils") OR human neutrophil antigens OR "HNA" OR human neutrophil antibodies) AND (testing OR assay OR detection OR determination OR kit OR kits OR typing OR genotyping OR polymerase chain reaction OR "PCR" OR "Enzyme-Linked Immunosorbent Assay" OR "ELISA") AND (blood Transfusion OR platelet refractoriness OR fetal/neonatal alloimmune neutropenia OR autoimmune neutropenia OR immune neutropenia OR alloimmune neutropenia OR "TRALI" OR "transfusion related acute lung injury" OR neonatal alloimmune neutropenia OR febrile non-hemolytic transfusion reaction OR post-engraftment neutropenia) AND (adverse OR risk OR complication OR safety OR side effect OR survival OR "false negative" OR "false positive" OR clinical outcome OR error OR incorrect OR mistyping OR invalid OR defect OR discordant OR accuracy OR success OR efficacy OR effective)

The search terms include the type of tests used to detect human neutrophil antigens and human neutrophil antibodies, as well as the diseases clinically impacted by HNA testing results such as TRALI, fetal/neonatal alloimmune neutropenia (FNAIN), febrile non-hemolytic transfusion reactions, post-engraftment neutropenia in hematopoietic stem cell transplantation, and autoimmune neutropenia.

We limited our literature search to full text articles published in English from January 1, 1994 to May 1, 2017. We believed that a review of the articles published in the last 23 years would cover the information needed to evaluate the analytical performance, safety and effectiveness of HNA devices. Conference abstracts were excluded from the literature review.

The search generated 355 publications. Titles and abstracts were reviewed to exclude studies not related to the safety, effectiveness or performance of FDA-cleared HNA devices. Studies conducted in the US or in other countries were included if they used FDA-cleared HNA devices. The literature does not typically use the term "FDA-cleared" when referring to the HNA assays used in studies. We compared the HNA assay name, type, and manufacturer information provided in each article with our database of cleared HNA devices, to determine whether the study used an FDA-cleared assay(s). Many studies referencing HNA typing were excluded as they did not specifically indicate the use of an FDA-cleared test (Figure 2).

A total of 335articles were excluded during the initial screening for the following reasons:

- Animal study
- Not specific to FDA-cleared HNA devices
- Not in English Language
- Inadequate analytical performance or clinical uses information

After titles and abstracts were reviewed, the full-texts of the remaining 20 articles were further examined for eligibility. A total of 14 articles were excluded during the full text review using the same criteria described above.
A total of 6 articles were included for systematic review. Additional information regarding the methodology for inclusion and exclusion criteria may be found in Figure 2.

2. Results

The results of our systematic literature review of FDA-cleared HPA and HNA devices to evaluate analytical performance and clinical uses are presented below. The literature reviewed consisted of HPA related studies (n=15), and HNA related studies (n=6).

3. Analytical Performance of FDA-cleared HPA Devices

In this section, we document that there were no published studies that evaluated the analytical performance of FDA-cleared HPA devices (typing or antibody detection).

3.1. HPA typing devices

The vast majority (14 out of 15) of the reviewed studies reported using nucleic acid-based technology to HPA type patients and donors in addition to using serological techniques; platelet immunofluorescence test (PIFT) and monoclonal immobilization of platelet antigen assay (MAIPA). However, the studies reviewed either did not discuss the analytical performances of the devices used or did not indicate whether the devices used were FDA- cleared.

The GTI ThromboType (HPA 1-6, 15) and ThromboType 1 (HPA1) are the only FDA-cleared genotyping assay kits intended for molecular determination of alleles of human platelet alloantigens HPA-1 ($P1^A$), HPA-2 (Ko), HPA-3 (Bak), HPA-4 (Pen), HPA-5 (Br), HPA-6 (Ca) and HPA-15 (Gov). The serological based typing assays such as: platelet immunofluorescence test (PIFT) and monoclonal antibody-specific immobilization of platelet antigens assay (MAIPA) are older techniques that are apparently being replaced by the genotyping assays.

3.2. HPA antibody detection devices

For platelet antibody detection assays, the majority of the reviewed literature reported using the FDA-cleared ELISA assays manufactured by GTI (PAKAUTO, PAK-2, MACE or PakPlus)[1,2,3,4,5,6,9,11,12,14,15]. Use of FDA-cleared Capture-P Ready assay from Immucor was also mentioned[3]. In addition, the serological method, monoclonal antibody immobilization of platelet antigens assay (MAIPA, non-FDA-cleared assay) was reported as being a confirmatory assay[18].

Like the HPA typing assay, the analytical performance of the HPA antibody assays was not discussed or mentioned in the literature reviewed. However, one article by Bessos, et al.[2] noted significant correlation between titer results of semi-quantitative GTI PAK 12 and antibody concentration by quantitative ELISA. The quantitative ELISA was developed internally to measure platelet antibody concentration in patients' serum. This study was conducted to assess the relationship between anti-HPA-1a concentration and severity of neonatal alloimmune thrombocytopenia (NAIT). The authors concluded that there is no relationship between anti-HPA-1a concentration and severity of NAIT when quantitative ELISA is used, and noted that the correlation between ELISA and other serological methods, remains to be determined.

Culler, el al.[3] reported finding anti-CD36 (anti-GPIV) in a 16-year old female with homozygous hemoglobin S disease evaluated for allogeneic hematopoietic stem cell transplantation (HSCT) through a routine pre-transplant platelet antibody screening. Platelet antibody screening was performed with GTI PAKPLUS. According to the PAKPLUS package insert cited in the study, the assay has co-positivity (sensitivity) of 88.2% and co-negativity (specificity) of 98.2% when compared with the reverse passive hemagglutination assay. HPA typing by flow cytometry revealed that the patient and her HLA identical brother lacked CD36. Platelet crossmatching using a solid-phase red cell adherence (SPRCA) assay, Immucor's Capture-P Solid Phase System, was also used to determine if sufficient quantities of compatible apheresis platelets were available to support the patient during HSCT. The patient received 49 units of apheresis platelets during the clinical course and experienced increases in platelet count following transfusion with crossmatch compatible platelets. The authors noted that this case illustrated the importance of routine screening of high-risk populations for platelet antibodies prior to requiring extensive platelet support.

4. Clinical Uses of HPA Testing Results to Aid Transfusion

HPA testing results have an important role in aiding the diagnosis and management of patients with fetal or neonatal alloimmune thrombocytopenia (FNAIT), platelet transfusion refractoriness (due to platelet alloimmunization) and post-transfusion purpura (PTP). HPA typing and/or antibody identification in patients with these conditions can enable effective platelet transfusion support during periods of thrombocytopenia.

4.1. Clinical uses of HPA results

FNAIT or FMAIT

In addition to using HPA results for platelet matching between donors and recipients, results from HPA have also been used in diagnosing disease. In suspected cases of fetal or neonatal allo-immune thrombocytopenia (FNAIT), initial diagnoses are usually based on "clinical grounds and depend on the exclusion of other causes of neonatal thrombocytopenia and maternal autoimmune disease which can cause

neonatal thrombocytopenia"[1]. The confirmation of FNAIT depends on finding maternal alloantibodies directed against antigens present on paternal platelets in a laboratory. The literature reviewed stated that 70% to 90% of the FNAIT cases in Caucasians can be attributed to antibodies against human platelet antigen HPA-1a followed by anti-HPA-5b[1, 2,4, 15, 18.] It was noted that "since the anti-HPA-1a antibodies are the major cause of Fetal-Maternal Allo-immune thrombocytopenia (FMAIT) and the incidence of FMAIT is considerably high, some authors suggest that HPA-1a phenotyping and antibody screening should become obligatory for all pregnant women within the pre-existing antenatal red cell serology programme"[16]. The terms FMAIT and FNAIT infer the same disease process.

Jeremiah et al.[12] performed a study investigating the frequency of alloantibodies to HPA antigens in Nigerian women with history of frequent pregnancies. They found that parity had a significant influence on the development to HPA antibodies. Using the GTI PakPlus qualitative solid-phase ELISA assay the prevalence of anti-HPA-5b was 30%, anti-HPA-5a 18%, and there were no antibodies to HPA-1a, -3a and -4a detected among the 100 Nigerian women tested. The authors indicated that although neonatal alloimmune thrombocytopenia (NAIT) has not been reported in Nigeria, it is possible that some cases of mild to moderate neonatal thrombocytopenia caused by anti-HPA-5b and -5a may have occurred but were not identified due to lack of platelet serology testing. They concluded that there is a need to establish a platelet serology laboratory for the proper antenatal and postnatal management of pregnant mothers in Nigeria.

Post-transfusion Thrombocytopenia

Katerina Pavenski et al.[14] reported using PCR genotyping kit, ThromboType from GTI (HPA-1 through -6 and -15) for genotyping of patient and donor to investigate unexplained post-transfusion thrombocytopenia following transfusion of fresh frozen plasma. Using ThromboType, the investigators determined the patient's genotype as HPA-1a/1b and donor's genotype as HPA-1b/1b. The donor tested positive for anti-HPA-1a. A lookback investigation associated the implicated donor with a previous case of post-transfusion thrombocytopenia. This case report of thrombocytopenia due to passive transfer of platelet antibodies demonstrates the usefulness of both HPA typing and HPA antibody detection devices.

Platelet Transfusion Refractoriness

To improve the treatment of platelet transfusion refractoriness particularly in patients needing long-term platelet transfusion support in China, Xia, et al.[20], established a platelet registry consisting of 864 platelet aphaeresis donors with known HLA and HPA phenotypes. To establish the platelet donor registry, the donors were typed for HLA-A, -B and HPA-1, -2, -3, -4 -5, -6 and -15 using the PCR-SSP method. The group reported that significantly higher platelet recovery (PPR) values were obtained

after transfusion with HLA-matched platelets compared with PPR values after random platelets.

Using the GTI PAKPLUS ELISA kit, flow cytometry and nucleotide sequencing, Xu et al.[19] examined the prevalence of CD36 (also known as GPIV) immunization in China. CD36 deficiency is known to be responsible for the production of anti-Nak^a antibodies in various clinical settings such as fetal/neonatal alloimmune thrombocytopenia (FNAIT), platelet transfusion refractoriness (PTR) and post-transfusion purpura (PTP). This study tested 998 healthy blood donors and found that 18 individuals failed to express CD36 on their platelets. In 5 out of 18 donors, no CD36 expression was detected on both platelets and monocytes; indicating the frequencies of type 1 CD36 deficiency (platelets and monocytes) and type II CD36 deficiency (platelets only) are approximately 0.5% and 1.3%, respectively. The investigators concluded that more than 0.5% of CD36 type 1-deficient individuals are at risk of immunization through blood transfusion or pregnancy in China and that testing for anti-Nak^a antibodies should be considered in suspected cases of FNAIT and PTR. In addition, a registry of CD36-deficient donors should be established to allow treatment of immune-mediated bleeding disorders caused by anti-Nak^a antibodies.

Friedman et al.[6] tested their hypothesis that antiplatelet antibodies would be detected frequently in transfused sickle cell disease (SCD) patients by screening the sera of heavily transfused, moderately transfused and un-transfused SCD patients for platelet antibodies utilizing Immucor's Capture-P Ready Screen assay. The assay was used according to the manufacturer's instructions for use. The study reported that both heavily and moderately transfused SCD patients have a high prevalence of platelet alloimmunization at85% and 48% respectively, compared to 0% in un-transfused SCD patients. The findings suggest that these patients could potentially exhibit significant platelet refractoriness, a complication sometimes experienced by sickle cell disease patients undergoing bone marrow transplantation (BMT).

5. **Analytical Performance of FDA-cleared HNA Devices**

The main limitation of our systematic literature review for HNA devices is that there are only two FDA- cleared HNA assays to detect granulocyte antibodies, and only two HNA assays to detect granulocyte antigens. Consequently, there is little published literature describing the analytical performance, safety and effectiveness of these FDA- cleared devices. Serological techniques such as Granulocyte Immunofluorescence test (GIFT), Granulocyte agglutination test (GAT) and monoclonal antibody-specific immobilization of granulocyte antigens (MAIGA) are not FDA-cleared devices.

5.1. HNA typing devices

In this literature review, we only considered articles with data generated from FDA-cleared HNA devices. In regards to the two FDA-cleared devices used to detect

human neutrophil antigens, we did not identify any publication that evaluated the two assays below.

- HNA Genotyping Tray (One Lambda): this assay is a PCR based molecular assay using sequence specific priming (SSP) for the detection of polymorphisms and to determine molecular alleles. This assay is used for the molecular determination of neutrophil polymorphisms HNA-1a, 1b, 1c, 3a, 3b, 4a, 4b, 5a, and 5b.

- GranType (GTI): this is a DNA based assay for the molecular determination of polymorphism HNA-1a (NA1), HNA-1b (NA2), and HNA-1c (SH).

5.2. HNA antibody detection devices

LABScreen Multi Assay from One Lambda, Inc., is the only HNA antibody detection device cleared by FDA. This assay is a Luminex-based antibody detection assay originally designed to detect antibodies to human HLA and HNA-1a, -1b, -1c, and -2. LabScreen Multi assay has been modified to also detect HNA-3a, -3b, -4, -5a, and -5b antibodies.

Fromont et al.[7] investigated 51 sera from cases with defined alloimmune profiles (i.e. from alloimmune neutropenia and TRALI cases) and 40 sera from autoimmune neutropenia cases. The samples were tested for granulocyte antibodies using One Lambda LABScreen Multi, and compared with results from classical tests such as flow cytometry (FC), and granulocyte agglutination (GAT) followed by monoclonal antibody-specific immobilization of granulocyte antigens (MAIGA). The authors used the LABScreen Multi with the manufacturer recommended cut off. The study showed that in alloimmune conditions (n = 51), 94% (48 sera) were concordant (kappa coefficient of 0.85, -attesting a very good concordance), 2 sera that were positive for HNA with LABScreen Multi were negative by FC/GAT and/or MAIGA (2 false positive samples), and one serum sample negative for HNA with LABScreen Multi was positive by classical tests (1 false negative sample). In autoimmune neutropenia (n = 40), 75% (30 sera) were concordant, 4 sera positive for HNA with LABScreen Multi were negative by FC/GAT and /or MAIGA, and 6 sera negative for HNA with LABScreen Multi were positive with FC/GAT and/or MAIGA. Except for one sample, the discrepancies were observed in sera that did not show a clear specificity. In autoimmune conditions, the One Lambda LABScreen Multi was less concordant with classic methods for detection of autoantibodies (kappa coefficient 0.49 = moderate concordance) and there was less agreement in terms of antibody specificity.

This study did not include comparison with well characterized clinical samples to reveal more information about the sensitivity and specificity of LABScreen Multi. For detection of alloantibodies directed against HNA-1 or HNA2 antigens, the LABScreen MULTI was considered a good alternative to GAT and FC since 94% of results were concordant, but use with caution was recommended for screening

of autoantibodies. The study concluded that HNA-3 antibodies (responsible for severe cases of TRALI) should continue to be screened using GAT, because at the time of their publication LABscreen Multi beads did not contain HNA-3a, limiting the usefulness of the assay as a screening test for blood donors. The authors also recommended that HNA antibodies detected by LABScreen Multi should be confirmed using reference methods (FC and MAIGA) with genotyped neutrophils.

- Heinzl et al.[10], reported screening 333 samples for HNA antibodies using a combination of granulocyte aggregation test (GAT) and granulocyte immunofluorescence test (GIFT) and comparing to flow cytometric white blood cell immunofluorescence test (Flow-WIFT). GAT/GIFT detected 77 (23.1%) positive samples while Flow-WIFT found 108 (32.4%) granulocyte-reactive samples. For the comparison of GIFT with Flow-WIFT, Cohen's k was at 0.682, signifying good concordance between the two methods. Between the three screening methods, GAT, GIFT and Flow-WIFT, 114 samples tested positive. Positive samples were further analyzed using MAIGA and One Lambda LABScreen Multi. Of the 114 positive samples, 70 were confirmed positive and 31 confirmed negative by both, MAIGA and LABScreen Multi; 9 detected as positive by LABScreen were negative by MAIGA; and 4 negative by LABScreen were positive by MAIGA. The correlation between MAIGA and LABScreen Multi was reported as Cohen's k = 0.742 .

  Although results between One Lambda LABScreen Multi and MAIGA were shown to correlate well, One Lambda Multi appeared to be less reliable than MAIGA for HNA-antibody specification. This was caused by questionable specification results based on observed panreactivity (positive with all beads) of microbeads in the LABScreen Multi but negative results by other screening methods; GAT, GIFT and Flow-WIFT. It was speculated that the questionable results occurred because the antigens on the microbeads are recombinant and might thus have an altered conformation and density in comparison with antigens expressed on natural cells.

- Schuz et al.[17] evaluated the new generation LabScreen Multi assay by comparing it to the first-generation LABScreen Multi assay. The study tested large pools of blood donors to mitigate the risk of TRALI by screening for HNA-1a, HNA-1b, and HNA-2 antibodies Among the 10,411 donors tested using the first-generation device, 666 (6.4%) had positive test results, which were predominantly non-specific. Only 33/666 (4.9%) of the positive LABScreen Multi results were confirmed with GIFT or GAT, corresponding to a total of 0.3% of all tested donors. The second generation LABScreen Multi assay contained additional beads for the detection of HNA-3a, HNA-3b, HNA-4a, HNA-5a, and HNA-5b antibodies which were lacking in the first-generation assay. To test the performance of the second-generation device, 97 sera samples containing well-defined HNA antibodies were used. In addition, 91sera from blood donors that had previously tested negative for HNA antibodies with the first-generation assay,

and were confirmed negative with GAT, GIFT and the monoclonal antibody-specific immobilization of granulocyte antigens (MAIGA) assay were tested using the second-generation device. The performance study showed that the second generation LABScreen Multi assay was highly specific for the HNA-1a, HNA-1b, HNA-2 and HNA-3a. The assay detected 98% of true positives for HNA-1a, HNA-1b and HNA-2 antibodies, and 90% of the true positive HNA-3a antibodies were recognized and identified correctly. False positives were identified in 5.5% of the 91 samples that were confirmed negative previously. The investigators stated that the detection of HNA-3a antibody specificities could be integrated into the new generation LABScreen MULTI assay; although, the specificity for HNA-3a is only 90%. In addition, they noted that other HNA antibodies were detected with the new generation assay, such as HNA-1c, HNA-1d, and some HNA-3b and HNA-4a antibodies. The investigators concluded that although, the new generation LABScreen MULTI is a great step toward feasible high-throughput testing for HNA antibodies, GIFT and GAT remain the gold-standard for differentiating rare and currently unknown HNA specificities.

6. Clinical Uses of HNA Testing Results to Aid Transfusion

6.1. Clinical uses of HNA results

The reviewed literature mainly discussed using HNA devices to investigate or prevent occurrence of transfusion-related acute lung injury (TRALI) from blood transfusion. TRALI is described as a severe complication of transfusion which may occur within 6 hours of transfusion. “Antibodies directed to human leukocyte antigens (HLAs) or human neutrophil antigens (HNAs) have been identified as causative agents in TRALI by observational studies and by the reproduction of antibody induced TRALI in an ex vivo lung model. Fatal TRALI reactions are often caused by HNA-3a and HLA Class II antibodies, which are mainly produced during pregnancy”. [17]

Schulz et al.[17], described using the second generation One Lambda LABScreen Multi Luminex bead assay to test for human leukocyte antigen and neutrophil antigen antibodies in a high number of plasma donors to reduce the risk of TRALI. They found that compared to the first generation One Lambda LABScreen Multi test, the second-generation assay was highly specific for the HNA-1a, HNA-1b, HNA-2 and HNA-3a antibody specificities.

Similar studies have also been conducted using the combination of first-generation One Lambda LabScreen Multi and serological methods (GIFT or GAT). Serological testing is included to detect other HNA antibodies not included in the first-generation LabScreen Multi assay particularly HNA-3 to reduce risks from TRALI[8,12,13].

Xia, et al.[21] utilized LabScreen Multi assay from One Lambda and an in-house ELISA assay to examine the prevalence of anti-human neutrophil antigen (anti-HNA) antibodies

against HNA-1a, -1b and -2 in blood donors from South China that are responsible for transfusion-related acute lung injury (TRALI). An antigen capture assay was used to detect anti-HNA-3, with confirmation by the granulocyte agglutination test (GAT). The investigators reported testing blood samples from 778 donors (randomly selected from 1014 donors) and identifying three donors with anti-HNA antibodies; two against HNA-2 and one without known specificity. The anti-HNA-2 specificity was confirmed by granulocyte immunofluorescence test (GIFT). Anti-HNA-3 antibodies were not found. The rate of alloimmunization against HNA was found to be 0.39% compared to 4.63% and 24.7%, for HLA classes I and II respectively, in the female donors that were tested. The investigators concluded that although immunization against HNA seems to be a rare event in China, further observation is necessary to determine the necessity of HNA antibody screening in Chinese blood donors.

In addition to TRALI, One Lambda Multi assay along with serological assays (GIFT, GAT, and MAIGA) have been used to detect antibodies directed against granulocyte-specific antigens that have been implicated in alloimmune and autoimmune neutropenia[7,10].

7. Safety and Effectiveness Associated with the Clinical Use of HPA and HNA Testing Devices

It appears that the FDA-cleared devices are largely effective in providing correct test results to aid in the diagnosis and treatment of certain diseases. However, HPA and HNA devices can malfunction. They can produce inaccurate test results (false positive and false negative); they can fail to correctly interpret test results (based on uncertainty around setting the appropriate cutoff value); some devices may have a low sensitivity, a low positive predictive value (PPV) or a low negative predictive value (NPV), or discrepancies may exist between phenotype and genotype determinations.

HPA devices and HNA devices are used to support the clinical diagnosis of several diseases. An inaccurate diagnosis due to incorrect test results could have clinical and therapeutic consequences with a negative impact on the patient. Some examples are described below.

Poles et al.[15] reported that low-frequency HPAs are clinically important in cases of fetomaternal alloimmune thrombocytopenia (FMAIT); however, these low-frequency HPAs are not presented in normal panel platelets or antibody detection and identification assays such as GTI PakPlus. Consequently, testing with these assays failed to detect HPA antibodies in the maternal serum. FMAIT is typically caused by the transfer of antibodies against fetal human platelet antigens inherited from the father that are absent in the mother. There antibodies result in fetal and neonatal thrombocytopenia and clinical implications range from intrauterine growth retardation to intracranial hemorrhage and fetal loss.

Goldman et al.[9] noted in their Report on the 11th International Society of Blood Transfusion Platelet Genotyping and Serology Workshop, that many laboratories combine more than one testing method in difficult neonatal alloimmune thrombocytopenia (NAIT) cases and almost all laboratories have encountered difficult cases which clinically appear to be NAIT, but no platelet alloantibody is readily detectable.

Fromont et al.[7], attempted to improve agreement between the One Lambda LABScreen Multi assay and classical method (e.g., MAIPA) by proposing a different cutoff for auto and alloantibodies and one for each HNA specificity, but reported that they did not improve the agreement by increasing or reducing the cutoff. However, a consensus from other laboratories using this test proposed higher cutoff values. Using a similar approach the investigators showed similar results except for one serum sample containing HNA-2a antibodies, which could not be detected using a higher cutoff.

Schulz et al.[17], notes the possibility that special steric requirements for antibody also may be the cause for nonreactivity with the recombinant HNA-1b molecule coated on the bead of One Lambda LabScreen Multi assay. They also stated that increasing the cutoff for the HNA-2 bead might solve the problem with false-positive results but it could also lead to missing weakly reactive sera.

8. Overall Literature Review Conclusions

a. Analytical performance of the HPA and HNA typing and antibody detection devices

Our literature review did not identify any studies that evaluated the analytical performance of FDA-cleared HPA typing or HPA antibody detection devices. Similarly, there were no studies that evaluated the analytical performance of the FDA-cleared HNA typing devices. However, our literature review included a small number of studies that evaluated the analytical performance of the only FDA-cleared antibody detection device.

Overall, the studies showed that FDA-cleared HNA antibody detection device performed comparably with flow cytometry, granulocyte agglutination (GAT), monoclonal antibody-specific immobilization of granulocyte antigens (MAIGA) and other classical non-FDA-cleared tests. Differences in performance were noted depending on whether the disease was mediated by an auto- or alloimmune process, with concordance appearing to be better for alloimmune conditions compared to autoimmune conditions. It was also noted that the FDA-cleared HNA antibody detection device was less reliable than GAT, GIFT and MAIGA at differentiating HNA antibody specificity.

b. Clinical use of HPA and HNA devices in transfusion, and disease diagnosis

Testing results from HPA devices have been widely used to diagnose disease in the setting of fetal or neonatal thrombocytopenia, and to select appropriate antigen-negative platelet products for transfusion. HPA devices are also used to evaluate the etiology of platelet refractoriness in patients who have been frequently transfused and for selection of appropriate products when platelet antibodies are present.

In the transfusion setting HNA devices are used to evaluate adverse reactions in transfusion recipients and to mitigate the risk of TRALI potentially caused by blood donors with HNA antibodies. Other less frequent clinical uses described in the literature include use in diagnosing neonatal alloimmune neutropenia, autoimmune neutropenia and drug-induced neutropenia.

c. Risks associated with HPA and HNA devices

While studies report overall acceptable performance of FDA-approved HPA and HNA devices and widespread clinical use, device malfunction can occur causing false positive or false negative results. Spurious results may affect patient outcomes through delayed or missed diagnoses, delayed treatment and selection of inappropriate or ineffective products for transfusion.

Discussions of Strengths and Limitations

This literature review was limited by the relatively small number of published studies that evaluated FDA-cleared HPA and HNA devices. There were several studies reporting on the use of HPA and HNA devices in select clinical populations. These studies provided important information for diagnosing and treating related conditions.

Conclusions

Based on our systematic review of the literature, we found that FDA-cleared HPA and HNA devices demonstrate acceptable analytical performance, are safe and effective, and play a critical role aiding in the diagnosis of autoimmune and alloimmune thrombocytopenic and neutropenic conditions and can also be useful in guiding selection of appropriate transfusion therapy when indicated.

Figure 1: Workflow of article selection described in Section 1.1

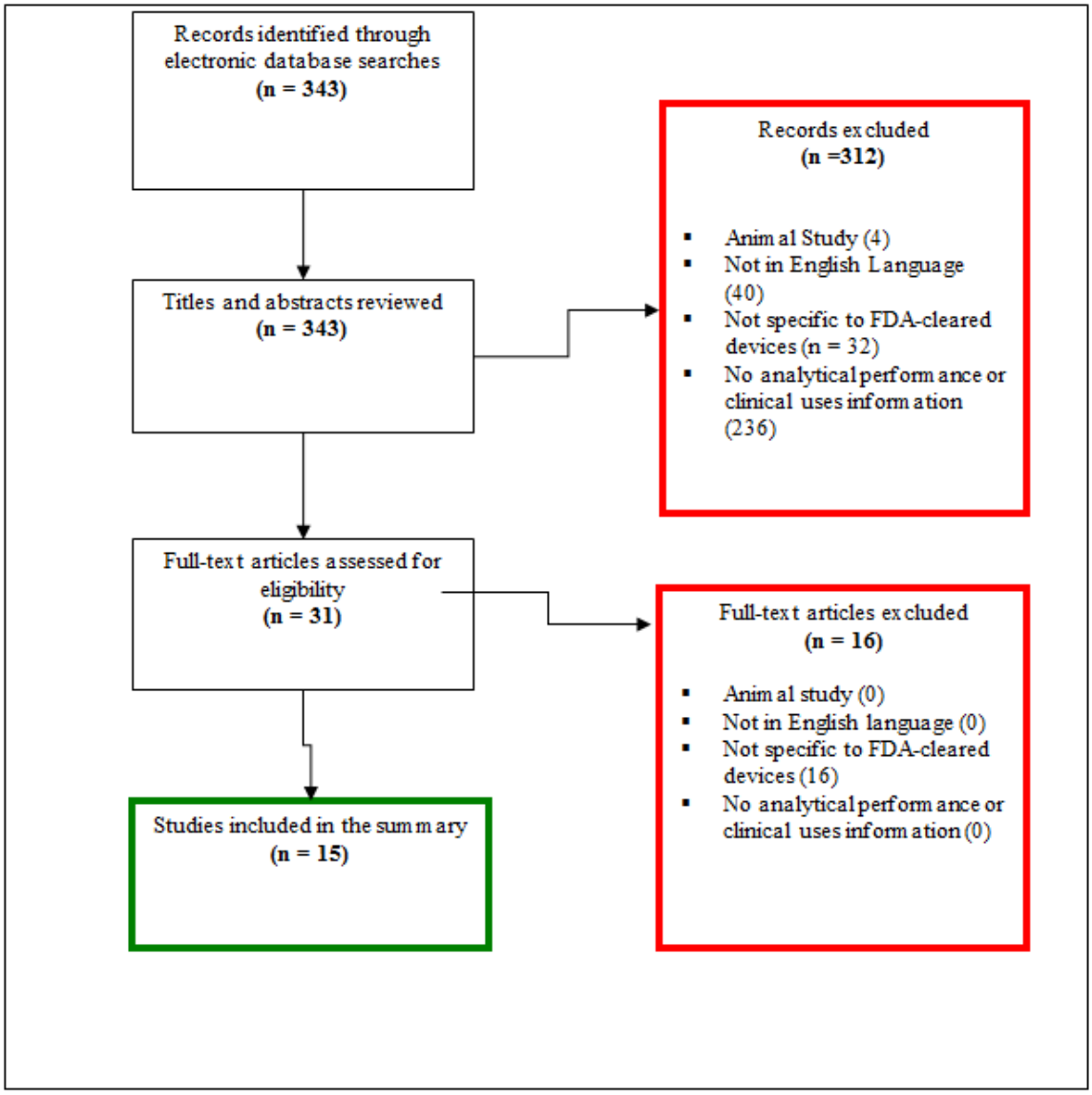
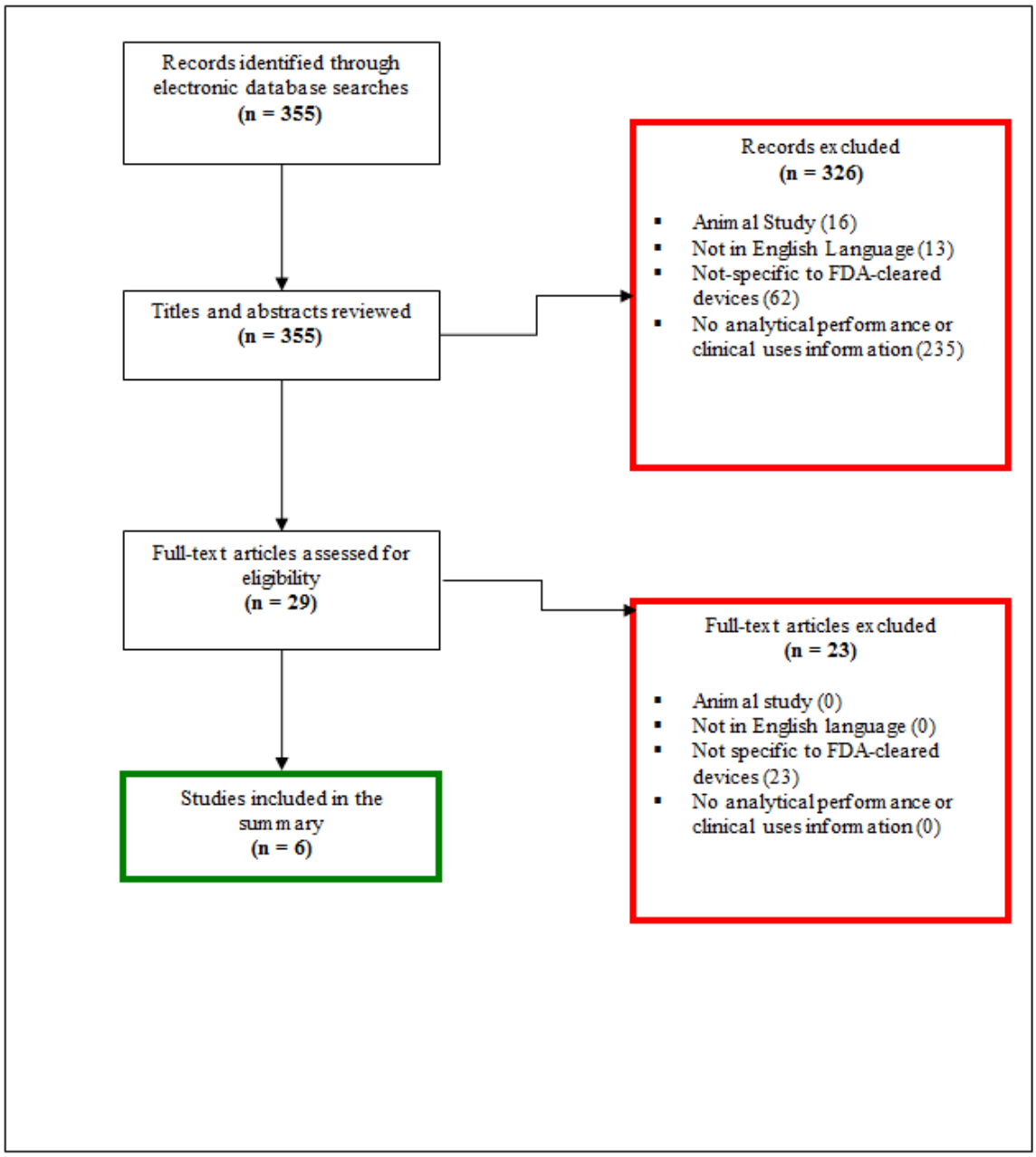


Figure 2: Workflow of article selection described in Section 1.2



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