

Heterophilic Antibody Interference in Laboratory Tests: Important for Clinicians and Practicing Pathologists

Deborah V. Spencer, MD

Naturally occurring heterophilic antibodies such as those that occur in infectious mononucleosis are known to cause interference in assays in the chemistry laboratory. These nonspecific antibody interactions can cause incorrect associations between patient sera and test assays, causing a false-positive or a falsely elevated test result. Heterophilic antibodies have caused falsely elevated results for assays such as human chorionic gonadotropin, prostate-specific antigen, and cancer antigen 125 (CA 125), causing negative outcomes for patients.¹ Heterophilic antibodies have been reported to cause a false-positive result in a rapid human immunodeficiency virus (HIV) test, illustrating that these antibodies can cause problems in point-of-care tests as well as laboratory assays.² Clinicians and pathologists need to be cognizant of the challenges caused by heterophilic antibodies so that appropriate steps may be taken to eliminate antibody interference in chemistry assays.

Heterophilic antibodies are common, naturally occurring antibodies with low affinities. These antibodies initiate weak interactions between antigens and antibodies, and may function to regulate the immune system. Medical researchers have proposed that heterophilic antibodies bind and remove foreign antigens from the intestinal tract and help maintain self-tolerance.³ These antibodies react with many antigens, including a wide variety of chemical structures, self-antigens, and the variable region of other antibodies (anti-idiotypic antibodies). Heterophilic antibodies are inherently produced from B cells prior to antigen exposure and result from random combinations of the genes of the heavy and light chain variable regions. Heterophilic antibodies are not the same as specific human anti-animal antibodies, which are produced when animal antibodies are injected into a patient for treatment or diagnostic purposes.^{4,5} Anti-animal antibodies are specific for antibodies produced by the particular animal, and they only interfere with assays using antibodies from that particular species.⁶

Many non-competitive laboratory assays including hormones, cardiac markers, tumor markers, and other serological tests such as for infectious disease testing are subject to heterophilic antibody interference, with potentially drastic consequences. Two-site (sandwich) laboratory assay protocols for detection of antibodies or antigens are susceptible to weak heterophilic antibody interference because there is little competition for binding.^{3,7} Heterophilic antibodies are typically not strong enough to interfere with competitive binding assays.⁷ Many heterophilic antibodies are specific for the antibody binding fragment (Fab) of other antibodies, and react with a specific set of assay antibodies. These heterophilic antibodies will cause interference with one assay, but they may react poorly with another assay's antibodies. Switching the assay kit to

one from a different manufacturer or to an in-house preparation may reduce or eliminate the interference by removing the idiotope with which the heterophilic antibody reacts. The use of nonimmune globulin or serum from several animal species or commercially available preparations such as heterophile blocking reagents (HBR) and immunoglobulin inhibiting reagents (IIR) can significantly reduce heterophile interference.^{1,3}

Clinicians need to be aware of the challenges caused by heterophilic antibodies because of the increasing use of point-of-care tests in clinical practice. Heterophilic antibody interference should be considered when weak false-positive or slightly elevated test results are encountered, especially if the test results are incongruent with the clinical status of the patient. Prompt communication between clinicians and the laboratory is essential so that a second alternate test may be used, and if necessary, heterophile blocking reagents (HBR) or immunoglobulin inhibiting reagents (IIR) used to investigate suspected interference, followed by confirmatory testing as appropriate and available. By working together, we can identify false-positive results caused by heterophilic antibodies and have a profound impact on the clinical management of patients.

References

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