1 Evolution of Veterinary Transfusion Medicine and Blood Banking

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Introduction

From ancient times to the modern day, knowledge of transfusion medicine and blood banking has advanced from blood existing as a spiritual fluid of vitality to it being a lifesaving therapeutic resource used on a regular basis. The most significant advancements in transfusion medicine have been made during the past 200 years, with veterinary transfusion medicine becoming a specialized area of interest for the past few decades. Transfusion medicine has progressed from fresh whole blood transfusions to targeted component therapy, with veterinary professionals performing transfusions in small, large, and exotic animals. Providing a safe and reliable blood product with availability that meets demands is now an emerging focus, as new knowledge cautions practitioners that transfusions, even when properly administered, can be harmful to patients.

Advancements in veterinary transfusion medicine include blood typing, compatibility testing, laboratory diagnostics to determine whether a transfusion is indicated, proper administration and dosage of blood products, as well as prevention, monitoring, and treatment of transfusion-associated complications. Veterinary blood banking has progressed from whole blood collection on an emergency basis with minimal regard to pre-transfusion compatibility testing, to the collection, storage, and processing of blood components and transfusion only after suitable recipient screening. This has led to the establishment of commercial blood banks and processing of blood products using specialized equipment, with evidence-based guidelines regarding donor screening. Additional advancements include methods to maximize the limited donor pool and awareness of storage lesions, as well as safety measures such as leukoreduction. Professional organizations such as the Veterinary Emergency and Critical Care Society (VECCS), American College of Veterinary Emergency and Critical Care (ACVECC), American College of Veterinary Internal Medicine (ACVIM), and American College of Veterinary Anesthesia and Analgesia (ACVAA), among others, actively pursue advancement of knowledge in the field of veterinary transfusion medicine and blood banking. Veterinary transfusion medicine as a specialty area of knowledge is growing, as seen through the re-emergence of efforts to establish sustainable organizations such as the International Association of Veterinary Blood Banks (IAVBB), the Association of Veterinary Hematology and Transfusion (AVHTM), and the proposed Academy of Veterinary Transfusion Medicine Technicians (AVTMT). Veterinary transfusion medicine is a discipline in its own right and will continue to play a vital role in veterinary medicine in an effort to improve patient care.

History of transfusion medicine

Ancient knowledge

Early practices and customs relating to the blood of ancient days include people drinking the blood of fallen gladiators to gain strength, religious figures attempting to heal themselves by drinking blood from the youth, and doctors inducing hemorrhage to let out "bad blood" due to the belief that blood was one of the four fundamental humors of Hippocratic medicine and blood-letting would bring balance to the humors and restore health (Greenwalt 1997). Early practices were often influenced by religion and superstition, as well as innate emotions and fears elicited by the sight of blood. People believed blood was the key to vitality, even though the discovery and description of the circulatory system did not occur until the 17th century.

Early concepts

It is unclear who first conceived the idea of blood transfusions. Hieronymus Cardanus (1505–1576) is given credit in some literature, while Magnus Pegelius obtained the right to publish on the topic under Emperor Rodolphus II's rule in 1593. Andreas Libavius was the first person clearly documented in history to advocate for blood transfusions; he recorded his thoughts on using a silver tube to connect the arteries of two individuals to allow blood from the young man to "pour" into the artery of the old man. However, there is no evidence indicating that transfusions were performed by Libavius (Greenwalt 1997).

Following William Harvey's description of the circulatory system, Francesco Folli of Florence published the first book on transfusions stating that transfusions could be used to treat illness and rejuvenate aged men. However, Folli stated in the book that that he had never performed a transfusion with the apparatus that he described was needed for the procedure (Greenwalt 1997).

Manual of Veterinary Transfusion Medicine and Blood Banking, First Edition. Edited by Kenichiro Yagi and Marie K. Holowaychuk.

Figure 1.1 A portrait of Richard Lower, a physician who performed the first reported animal-to-animal transfusion. (Public domain.)

First animal-to-animal transfusion

Richard Lower (1631–1691) performed the first successful animal-to-animal transfusion in February 1665; previous to this he had years of failed attempts due to clotting in the tubes (Figure 1.1). Lower used a medium-sized dog and exsanguinated it until "its strength was nearly gone", and then connected the cervical arteries of two large mastiffs to the jugular vein of the exsanguinated dog. The recipient in the experiment was "apparently oblivious to its hurts" and "soon began to fondle its master and to roll on the grass to clean itself of blood", indicating his first successful attempt to use a blood transfusion as a form of resuscitation. While Lower's report was published in 1666, Jean-Baptiste Denis (1635–1704) also claimed to have performed the first successful animal-to-animal transfusion; unfortunately, his report was delayed from publication for a year due to the imprisonment of the editor of the publication (Greenwalt 1997).

First animal-to-human transfusions

While similar uncertain claims to the first human transfusion have been made, Jean-Baptiste Denis is believed to have performed the first animal-to-human transfusions. He performed a transfusion of lamb blood to a 15-year-old child who was suffering from a persistent fever; the child was reported to have "a clear and smiling countenance" after the transfusion. Denis also performed a transfusion to the son of the Prime Minister of Sweden (Baron Bond), without successfully curing him, and to others without complications (Greenwalt 1997).

Lower, who had performed the first animal-to-animal transfusion, also performed an animal-to-human transfusion in 1667 to

Figure 1.2 A depiction of an animal-to-human transfusion performed in the 1600s. (Wellcome Library, London. Boutesteyn Leyden 1692. Creative Commons.)

Arthur Coga, who was described as a "harmless lunatic" and "eccentric scholar" at Pembroke College. He received a transfusion from the artery of a sheep and was reported to have "found himself well" afterwards.

The most notable report of an animal-to-human transfusion was on 19 December 1667, when Denis treated a patient named Antoine Mauroy, a 34-year-old newlywed husband who ran away to Paris to spend time indulging in sensual pleasures (Figure 1.2). Denis thought that a transfusion of calf blood would help calm Mauroy's urges due to the gentle nature of calves. The transfusion was reported to improve Mauroy's issues, making him quieter. The procedure was repeated several days later, but that time Mauroy experienced burning in his arm, pain over his kidneys, and tightness in his chest. A day later, he exhibited bleeding from his nose and dark urine. This signifies the first report of a severe transfusion reaction, likely acute hemolysis. Mauroy's wife insisted that Mauroy be treated a third time 2 months later when he was exhibiting similar behavior, but Mauroy did not comply. He died the following night without receiving the transfusion. Mauroy's wife was bribed by Denis' enemies to state that a transfusion killed her husband, leading to Denis' trial for manslaughter, for which he was exonerated. Rumors suggest that Mauroy's wife poisoned him with arsenic, although the truth is unknown (Farr 1980).

Because of Denis' experiences in France, his enemies were able to instate the Edict of Châtelet, effectively banning transfusion practices in France. It is likely that the magistrates in Rome and the Royal Society also enacted similar bans, therefore while some experimental transfusions were performed in other parts of the world, advancements in transfusion medicine were halted for the next 150 years (Greenwalt 1997).

18th and 19th centuries

During the 18th century, the value of transfusions in patients with severe wounds and hemorrhage was revealed. In 1749, a member

Figure 1.3 A portrait of James Blundell, a physician who performed the first reported human-to-human transfusion. (Public domain: The National Portrait Gallery, Volume II, 1820.)

of the Faculty of Paris named Cantwell stated that transfusions should not be forbidden in desperate situations. In 1788, Michele Rosa published is findings that animals in severe shock required whole blood instead of serum for successful resuscitation.

During the 19th century, James Blundell (1790–1877), who had witnessed many women die from postpartum hemorrhage, performed experiments with animals in preparation for transfusions to his patients (Figure 1.3). He limited his patients receiving transfusions to those suffering from severe hemorrhage and applied the knowledge gained by John Leacock on the apparent harm of xenotransfusions (transfusion of blood from a different species), thus attempting human-to-human transfusions. While the archives are somewhat contradictory regarding the number of successful cases, records show that in 1829 Blundell was able to successfully save a 25-year-old woman with postpartum hemorrhage by transfusing blood from one of the surgical team members. The blood transfusion was performed with a brass syringe, although Blundell later developed an instrument called the "impellor", a funnel-like apparatus that was used well into the late 19th century (Figure 1.4). While Blundell voiced his opinion against the transfusion of animal blood to human patients, the practice remained prevalent as transfusion therapy returned to medical practice. However, reports of transfusions were rare, likely due to the fact that blood clotting was a common limitation in performing transfusions (Greenwalt 1997).

Blood groups discovered

In the late 1800s there was significant work done by various physicians to study the effects of transfusions between different species. In 1874, Ponfick presented his findings of residues from lysed red blood cells (RBCs) in a patient who died after receiving a transfusion from a sheep. Ponfick also observed detrimental physical effects including respiratory distress, defecation, and convulsions, as well as post-mortem findings such as dilated hearts, pulmonary and serosal hemorrhage, enlarged and congested kidneys, and hemorrhage of the liver in dogs, cats, and rabbits receiving sheep blood. Ponfick also described

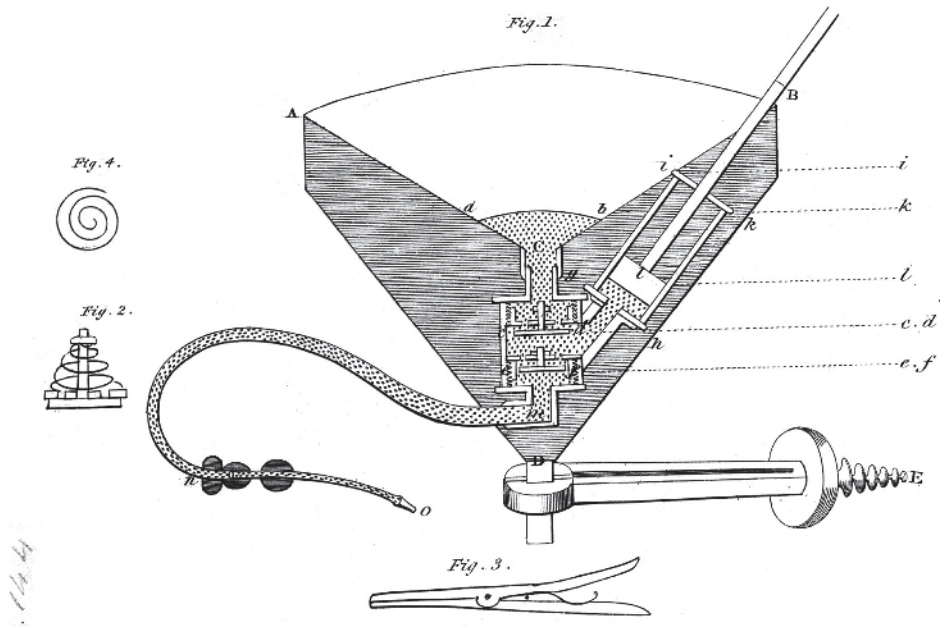

Figure 1.4 A section of the impellor device developed by James Blundell for blood transfusions. (Wellcome Library, London. Creative Commons.)

the accumulation of hematin in the renal tubules of surviving animals that developed kidney insufficiency. Ponfick's findings were consistent with Panum, Landois, and Euhlenberg's findings suggesting that adverse outcomes could be seen with transfusions between different species, secondary to hemolysis, kidney injury, and hyperkalemia (Greenwalt 1997).

In the 1800s, human-to-human transfusions were performed with a reasonable degree of success, frequently without signs of adverse reactions. This is probably because ABO incompatibilities in the general Caucasian population were only anticipated in one-third (35.6%) of randomly paired individuals (Greenwalt 1997). Nevertheless, there were still significant numbers of human-to-human transfusions resulting in fatal complications, which could not be explained by the work of Ponfick and others investigating inter-species transfusions (Greenwalt 1997). It was not until Landsteiner demonstrated agglutination using the serum from healthy humans mixed with another human's blood that the concept of blood groups (A, AB, B, and O) was established, which led to advancements in compatibility testing using assessments for agglutination (Landsteiner 1961). In 1910, von Dungern and Hirszfeld published a report on the inherited nature of blood groups; the practice of exclusively using O donors for transfusions began in the 1930s (Greenwalt 1997).

Advent of anticoagulation

The impellor was the tool designed by Blundell and used for transfusions until the 20th century. Another cannula device was devised by Crile in an effort to prevent blood clotting; it enabled the temporary joining of the recipient's vein and donor's artery, although it took significant surgical skill and strong donor will to accomplish this procedure. Other methods of transfusion included using paraffin to line the blood collection container, defibrinating the blood, and transfusing the non-clotted portion of blood (Greenwalt 1997).

Various anticoagulants were also studied in an effort to make the transfusion process more feasible, including the use of sodium phosphate by the well-known Braxton-Hicks, but none of his four patients receiving transfusions survived. Ammonium sulfate, sodium bicarbonate, sulfarsenol, ammonium oxalate, arsphenamine, sodium iodide, sodium sulfate, and hirudin (extracted from leeches) were all anticoagulant compounds investigated and reported by various physicians in the 19th and 20th centuries. In 1890, Nicolas Maurice Arthus reported that sodium citrate was able to permanently keep blood in liquid form, but it was not until 1915 that the invention of sodium citrate for blood transfusion was officially claimed. In 1955, Lewisohn was awarded the American Association of Blood Banks (AABB) Landsteiner Award for producing the first sodium citrate solution in a vial. Citrate was initially blamed as a cause of febrile non-hemolytic transfusion reactions, which were later determined to be the result of endotoxin from bacterial contamination (Greenwalt 1997).

Concept of blood banking

While blood mixed solely with 3.8% sodium citrate exhibited hemolysis after 1 week of storage, a mixture of blood, sodium citrate, and dextrose did not demonstrate hemolysis for 4 weeks. During World War I, Oswald H. Robertson established the first blood bank at the United States Army Base Hospital No.5 by using collection sets that were autoclaved and designed to collect up to 800 mL of blood into 160 mL of 3.8% sodium citrate. In 1937, an article written by Bernard Fantus at the Cook County Hospital in Chicago describes collecting 500 mL of blood into 70 mL of 2.5% sodium citrate into a chilled flask, then storing it under refrigeration at 4–6 °C. This became known as the first blood bank, which stored blood for 4–5 days (McCullough 2012).

While dextrose solutions were known to increase the storage time of RBCs, maintaining sterility was still an issue due to caramelizing of the dextrose solution during autoclaving of the collection system. In the 1940s, acid-citrate dextrose (ACD) solutions were developed; the addition of acidic forms of sodium citrate prevented caramelization, which allowed extension of storage of RBC products to 21 days (Greenwalt 1997).

As the potential storage time for RBCs increased, concerns regarding RBC metabolism during storage arose. It was already recognized that 2,3-diphosphoglycerate (2,3-DPG) was a substance present in RBCs, even though its role in oxygen binding was not yet elucidated. The level of 2,3-DPG was also observed to be lower in more acidic environments, leading to the development of citrate-phosphate-dextrose (CPD) solutions in 1947. These solutions raised the pH to 5.6 and the addition of phosphate resulted in better preservation of 2,3-DPG. By 1960, the introduction of additive solutions containing adenine increased the storage time (Nakao *et al*. 1960) and the RBC survival time was extended to 42 days (Simon *et al*. 1962). This vastly improved the ability to store RBCs instead of using fresh whole blood.

Plasma component use

The introduction of plasma component therapy occurred during World War II, mainly for the treatment of shock. Edwin J. Cohn and his colleagues developed the method of fractionation, thus enabling the use of human albumin and plasma as resuscitation fluids. Cohn's methods continue to be used today, with some modifications (Greenwalt 1997).

Invention of plastic bags and component processing

The patent for plastic containers for blood component therapy was filed by Carl Walter in 1950, which led to the development of component separation and transfusions that otherwise would not have been possible. The American Red Cross Blood Program experienced an increase in the use of packed red blood cells (PRBCs) from 0.8% to 88% of reported transfusions between 1967 and 1978 with the implementation of multi-chambered plastic bags connected by tubing (Greenwalt 1997). Baxter Corporation commercialized the invention with the Fenwal division (named partly after "Wal"ter), which later became its own company. The ability to separate plasma from RBCs led to the abundant supply of plasma and production of plasma protein concentrates, as well as the ability to produce platelet concentrates.

Plasma protein concentrates

In 1965, Judith Pool discovered that fresh frozen plasma (FFP) thawed at refrigeration temperatures would allow coagulation factor VIII to remain precipitated, leading to the administration of high concentrations of factor VIII to hemophilia patients during cryoprecipitate transfusions (Pool and Shannon 1965). In addition, Edwin Cohn developed the technique of creating factor VIII concentrates through fractionation, allowing for home storage of factor VIII in refrigerators and self-administration of factor VIII by hemophilia patients.

Platelets

The advent of multi-chambered plastic bags allowed for the separation of platelets into concentrates. The National Cancer Institute played a major role in investigating the use of platelet concentrates for the treatment of thrombocytopenia during the 1960s (McCullough 2012). Methods of preparing platelet concentrates and performing transfusions were established and reduced mortality rates in oncology patients with thrombocytopenia. The lifespan of platelet concentrates was initially a limitation as they were only viable for several hours, although Murphy and Garner established that they could be stored for several days at room temperature, which vastly improved the ability of platelets to be used as a transfusion product (Murphy and Gardner 1969).

Apheresis

Jack Latham developed the concept of separating blood components and selectively extracting the portions necessary for treatment, and established a semi-automated system for plasmapheresis (McCullough 2003). More recent improvements have allowed the separation and extraction of platelets, as well as leukocytes. Plasmapheresis is currently being investigated for its ability to remove antibodies and toxins (Crump and Seshadri 2009; Khorzad *et al*. 2011; Nakamura *et al*. 2012). Plateletpheresis continues to be a method of collection for platelet concentrates.

Leukoreduction

As fractionation of components into RBCs, platelets, and plasma became more common, the white blood cells (WBCs) that remained were considered residual in nature. WBCs cause febrile non-hemolytic reactions, transfusion-related immunomodulation, and can aid the transmission of specific viruses (Zimring *et al*. 2009). In the 1980s, methods of filtration by passing collected blood through a membrane were developed and termed "filter leukoreduction". This method is used in the majority of human blood banks today to reduce transfusion-related complications. Development of apheresis also led to the harvesting of components that do not contain leukocytes and is termed "process leukoreduction" (Zimring 2009).

The veterinary field

While the first experimental animal-to-animal transfusion was performed prior to transfusions between animals and humans, the development of veterinary transfusion medicine and blood banking is relatively recent. The first commercial veterinary blood banks were established in the late 1980s and more blood banks exist now than ever before. Many of the same concepts found in human transfusion medicine are employed in the veterinary field, with progressively larger numbers of veterinary studies being performed and findings presented to refine the practice of veterinary transfusion medicine.

Current veterinary transfusion and blood banking practices

Despite how common the practice of administering blood products has become in veterinary clinics worldwide, there is a remarkable lack of information regarding the transfusion practices used. While studies have been published documenting transfusion-related complications such as transfusion reactions, organ injury, or coagulopathies, little has been described in the literature as to how veterinary professionals are actually administering blood products or taking steps to ameliorate the consequences of transfusions. Comparatively, even less information is available describing the current use of veterinary blood donors. The little veterinary information published in this regard is in the form of surveys. While these surveys have selection bias and do not represent the views of the entire veterinary field, they function to provide some insight as to current veterinary transfusion practices.

Surveys on veterinary transfusion medicine and blood banking

The first survey documenting transfusion practices was published more than 20 years ago and included responses from 25 small animal clinics geographically stratified across the United States. It was a telephone survey that asked questions to exclusively small animal practices performing at least six canine blood transfusions per year. The survey responses revealed that the primary source of donor blood was from a "borrowed dog" at 48% of practices, an "in-house dog kept on the premises" at 48% of practices, and a "nearby veterinary school" at one practice. Two-thirds of practices performed infectious disease screening of blood donors and evaluated hematologic variables prior to donation, but only one-third determined the donor blood type. None of the practices reported blood typing recipients, but this survey was performed prior to the availability of in-hospital dog erythrocyte antigen (DEA) 1 blood-type tests. Approximately half of the practices surveyed did not recover the costs of the transfusion, which was considered a "lifesaving measure" in 80% of cases (Howard *et al*. 1992).

Two decades later, a web-based survey was performed, which compiled information regarding blood donor and transfusion practices from 20 veterinary teaching hospitals and 53 private referral hospitals located in the United States, Canada, Europe, and Australia. This survey reflects the practice of a select number of specialty hospitals performing blood transfusions, as only emergency and critical care or internal medicine specialists (not general practitioners) were surveyed (Jagodich and Holowaychuk 2016). However, the information collected provides an idea of what the current transfusion and blood banking practices are amongst some veterinary hospitals worldwide, demonstrating

how much transfusion practices have changed since the previous survey, performed more than 20 years earlier.

Current veterinary transfusion practices

The survey performed in 2012 provides information on transfusion practices used in specialty veterinary hospitals with regards to the blood products stored and/or administered, as well as recipient screening. PRBCs and FFP were the most frequently reported canine and feline blood products routinely purchased or collected by hospitals (Table 1.1), confirming a shift in transfusion practice from the collection and administration of whole blood to the routine use of component blood products (Jagodich and Holowaychuk 2016). This is in stark contrast to earlier transfusion practices as only 16% of previously surveyed small animal hospitals reported separating canine whole blood into components (Howard *et al*. 1992). Likewise, 96% of hospitals reported blood typing or crossmatching canine and feline recipients prior to blood product administration (Jagodich and Holowaychuk 2016), which is likely a reflection of the increase in knowledge and understanding of safe transfusion practices, as well as the availability of cage-side blood type kits, which were not available decades prior when routine recipient typing was not performed (Howard *et al*. 1992).

Current veterinary blood banking practices

The 2012 survey also provides information regarding the blood banking practices used in specialty veterinary hospitals, specifically concerning blood donor selection and screening. Approximately 50% of respondents reported using a combination of purchased blood products and hospital-run blood donor

Table 1.1 Percentage of surveyed hospitals that reported how frequently they purchased or collected different canine and feline blood products (Jagodich and Holowaychuk 2016).

Purchased	**Canine**				**Feline**			
Blood product	**Never**	**Rarely**	**Occasionally**	**Routinely**	**Never**	**Rarely**	**Occasionally**	**Routinely**
FWB	45	55	0	0	59	29	6	6
SWB	90	5	5	0	65	18	18	0
PRBC	0	0	0	100	6	0	18	82
FFP	0	0	0	100	6	0	29	71
CP	25	45	25	5	–	–	–	–
PC	40	45	5	10	–	–	–	–
PRP	45	35	15	5	–	–	–	–
LCP	60	25	10	5	–	–	–	–
Lalb	55	25	25	0	–	–	–	–
HBOC	70	25	0	5	82	18	0	0
Collected	**Canine**				**Feline**			
Blood product	**Never**	**Rarely**	**Occasionally**	**Routinely**	**Never**	**Rarely**	**Occasionally**	**Routinely**
FWB	5	25	42	28	0	25	20	55
SWB	47	21	21	11	63	9	9	19
PRBC	40	2	6	52	66	6	6	22
FFP	42	2	6	50	67	6	6	21
CP	77	11	6	6	–	–	–	–
CPP	77	13	4	6				
PC	40	45	5	10	–	–	–	–
PRP	70	13	15	2	–	–	–	–

CP, cryoprecipitate; CPP, cryopoor plasma; FFP, fresh frozen plasma; FWB, fresh whole blood; HBOC, hemoglobin-based oxygen carrier; Lalb, lyophilized albumin; LCP, lyophilized cryoprecipitate; PC, platelet concentrate; PRBC, packed red blood cells; PRP, platelet-rich plasma; SWB, stored whole blood.

programs to provide canine blood products, whereas 19% of hospitals provided canine blood products using hospital-run blood donor programs only. The majority (85%) of those hospitals reported routinely using staff-owned dogs as blood donors with fewer respondents (53%) using client-owned dogs. Only 11% of hospitals reported having a colony of canine donors in the hospital (Jagodich and Holowaychuk 2016). These results differ substantially from previously reported practices, which rarely purchased blood products and more commonly used in-house dogs (Howard *et al*. 1992). The change over the years is likely due to the development of commercial blood banks and a shift in ethical beliefs regarding keeping in-hospital colonies of donor dogs.

Infectious disease screening of canine blood donors was routinely performed at 94% of hospitals with a hospital-run blood donor program and 53% reported blood typing canine donors for DEA 1 (Jagodich and Holowaychuk 2016). This also represents an increase in diligent blood donor screening compared to that which was reported previously, likely due to an improvement in knowledge and understanding regarding safe transfusion practices.

While feline blood donor practices have not been previously reported, the survey performed revealed that similar to dogs, half of all hospitals obtained blood products from a combination of purchased blood products and hospital-run blood donor programs, whereas 26% reported obtaining feline blood products using only a hospital-run blood donor program. Staff-owned cats were used by 73% of hospitals, compared to 40% of hospitals that reported having a colony of feline donors and 36% using client-owned cats. Routine screening of feline blood donors for infectious diseases was reported by 98% of survey respondents (Jagodich and Holowaychuk 2016). These findings demonstrate a slight difference in thought with regards to using colony feline versus canine donors, but a high diligence with regards to enforcing safe transfusion practices.

Advancements in veterinary transfusion medicine

Several advancements have been made in the field of veterinary transfusion medicine during recent years and will continue to be made as more well-designed research studies are published. A PubMed search using the terms "transfusion", "veterinary", and "dog or cat" yielded 426 publications in the field of small animal transfusion medicine between 1965 and 2015 (Figure 1.5). Of these publications, 161 were published within the last 10 years. It seems that whereas studies used to be sparse, articles pertaining to veterinary transfusion medicine are now being published on a routine basis. Likewise, there has been a shift towards more prospective studies rather than case reports or retrospective investigations. All of these publications have served to enhance knowledge in the field of veterinary transfusion medicine and encourage an evidence-based approach to transfusion practices.

Evidence-based guidelines

The evidence-based approach to formulating veterinary transfusion guidelines has culminated in the publication of a consensus statement by the ACVIM regarding blood donor screening. This

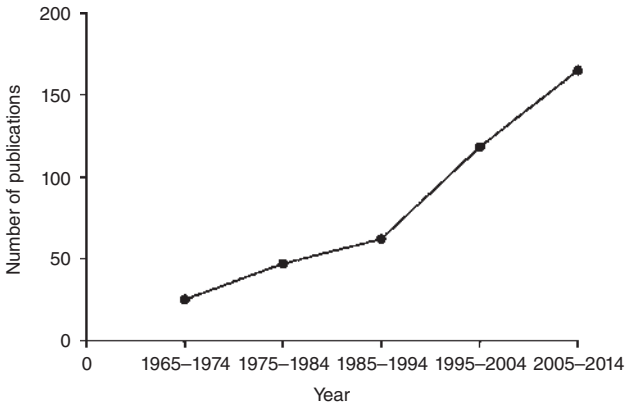

Figure 1.5 Graphical depiction of the number of veterinary publications related to transfusion medicine in dogs or cats.

consensus statement was drafted by a group of experts in the field of veterinary infectious disease and blood banking, and was first published more than 10 years ago (Wardrop *et al*. 2005). As a testament to the quickly growing body of research in the field of transfusion medicine, these guidelines were re-drafted and a preliminary view was provided at the ACVIM Forum in June 2015. The final recommendations were not published at the time of writing, but are anticipated to be published in 2016. Changes will likely reflect our increasing knowledge of infectious disease, including adjusted screening for feline leukemia virus (i.e., proviral DNA PCR testing) in cats, as well as banking samples from donors to allow retroactive testing.

The IAVBB is in the process of drafting and publishing veterinary blood banking standards modeled after guidelines provided by the AABB in the human field. These guidelines are expected to cover important details regarding the operation of a veterinary blood bank, such as the organizational structure, blood banking resources, equipment standards, supplier and customer issues, process control and improvement, documentation, facility standards, and safety. Without a doubt these guidelines will be the first of many to be published guiding veterinary transfusion and blood banking practices in the future.

Blood typing and recipient screening

Several advancements have also been made with regards to blood typing and recipient screening in dogs and cats. Whereas blood typing was previously only available at commercial laboratories and almost never performed at veterinary hospitals, the use of in-hospital blood type tests has become commonplace. This has served to improve the safety of blood transfusions administered in veterinary practice and likely has also enhanced the comfort level of practitioners administering blood products. Continued developments in this field have also improved typing methods, resulting in the availability of new canine and feline blood typing cartridges that use immunochromatographic test strips. Unlike agglutination card tests, the results of immunochromatographic tests can be interpreted even when auto-agglutination is present (Seth *et al*. 2012).

Other advancements in the field of blood typing include the discovery of new RBC antigens, including canine Dal and feline Mik (Blais *et al*. 2007; Weinstein *et al*. 2007). The detection of these antigens has changed recommendations with regards to donor and recipient screening, given that these antigens are not tested for by conventional blood typing methods. As such, some believe that all dogs and cats should routinely have a crossmatch performed prior to transfusions in order to maximize the potential to detect any incompatibilities not detected by conventional blood typing methods. This recommendation is emphasized by a recent study determining that feline red cell transfusion recipients that were blood type and crossmatch compatible had a higher post-transfusion increase in packed cell volume, compared to cats that were not crossmatched (Weltman *et al*. 2014).

The nomenclature of canine blood types has also recently changed, as it was discovered using flow cytometry that the DEA 1.2 and 1.3 blood types, which were previously thought to be different alleles, are likely a variation in the strength of monoclonal antibodies to DEA 1.1 (Acierno *et al*. 2014). Therefore, the nomenclature of DEA 1.1, 1.2, and 1.3 has become obsolete and is now described simply as DEA 1. This has already been reflected in a blood-type kit manufacturer's decision to rename the kit DEA 1, previously DEA 1.1 (DEA 1 Quick Test, Alvedia, France).

Transfusion triggers

Modification of the traditional transfusion triggers of 30/10 (packed cell volume 30%/hemoglobin 10 g/dL [100 g/L]) has occurred in human transfusion medicine in light of a multitude of studies demonstrating that a more conservative transfusion strategy (i.e., transfusing at a lower hemoglobin) is equal, if not superior, to the traditional and more liberal transfusion strategies (Carless *et al*. 2010). While research into the use of transfusion triggers is lacking in veterinary medicine, a scoring system has been developed to assist veterinarians in determining when a RBC transfusion might be warranted in anemic dogs (Kisielewicz *et al*. 2014). This score will likely guide veterinarians with less experience giving transfusions to more objectively determine when a transfusion might be warranted and also function to stratify patients being enrolled in future prospective transfusion studies.

Storage and administration of blood products

A relatively large number of studies investigating the effect of storage conditions and administration methods on the viability of veterinary blood products have been published in recent years. These include studies investigating various freeze-thaw conditions and storage temperatures on the activity of clotting factors in canine plasma products (Yaxley *et al*. 2010; Grochowsky *et al*. 2014; Walton *et al*. 2014; Pashmakova *et al*. 2015), as well as the impact of syringe or fluid pump administration methods on red blood cell viability (McDevitt *et al*. 2011; Heikes and Ruaux 2014). These studies, while experimental in nature, have improved our knowledge and understanding of the potential impact of storage, thawing, and administration methods on blood product viability and have immediate potential for clinical application.

Storage lesions and leukoreduction

Interest in storage lesions and the impact of the age of blood products on patient morbidity and mortality has recently increased (Obrador *et al*. 2015), along with research investigating the beneficial effects of pre-storage leukoreduction (McMichael *et al*. 2010; Graf *et al*. 2012; Herring *et al*. 2013; Corsi *et al*. 2014; Smith *et al*. 2015). There are also veterinary studies documenting the negative impact of administering older stored blood compared to blood stored for a shorter duration of time (Hann *et al*. 2014), while a clinical reduction in adverse effects associated with the use of leukoreduction filters has yet to be documented. As such, despite the relatively widespread use of leukoreduction in human medicine, routine use remains rare in veterinary medicine (Jagodich and Holowaychuk 2016). Likewise, the delineation of "fresh" versus "old" stored blood products is wrought with problems, including the increased disposal of expired blood products not used due to the negative connotations of stored red cell products (Holowaychuk and Musulin 2015). More information is needed with regards to the impact of storage lesions and leukoreduction on transfusion-related complications before firm recommendations can be made.

Therapies to reduce allogenic transfusions

Even though veterinarians are administering transfusions as safely as possible by performing diligent donor and recipient screening, and using appropriate administration and monitoring protocols, there is a growing concern regarding complications such as transfusion-related immunomodulation occurring secondary to allogenic transfusions (Hart *et al*. 2015). This has led to reports describing methods to reduce the administration of allogenic blood products. Examples include the use of specialized equipment such as cell salvage devices to enable safe and efficient autotransfusion of body cavity hemorrhage (Kellett-Gregory *et al*. 2013), as well as the administration of antifibrinolytic medication to ameliorate post-operative hemorrhage and transfusion requirements in predisposed breeds such as greyhounds (Marin *et al*. 2012a,b). It is likely that studies focused on reducing allogenic transfusions will continue to be performed as veterinarians seek out alternatives.

Future directions

Even though the number of veterinary studies published in the field of transfusion medicine is rapidly growing, there is still much work to be done and more knowledge to be gained in order to guide transfusion and blood banking practices. While retrospective studies have documented transfusion-related complications and demonstrated their association with a negative outcome, prospective studies are needed to further characterize what can be done to ameliorate these complications. Whether this will mean changing donor and recipient screening, adjusting transfusion triggers, using leukoreduction filters, altering blood storage and administration protocols, or seeking alternatives to allogenic transfusions remain to be determined.

Sourcing of sufficient donors to meet blood bank demands is also a consistent issue. Efforts to create wider public awareness of the need for donors, find an effective and sustainable supply of donated blood products, and use alternatives such as hemoglobin-based oxygen-carrying solutions and stem-cell derived RBCs, in addition to further refinement and widespread education regarding the appropriate use of blood products should help meet blood product demands. There is no doubt that the coming years will bring a plethora of veterinary publications that will serve to enhance knowledge and understanding of transfusion medicine and blood banking, enabling the creation of more evidence-based guidelines.

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