Commissioned Review Article

Feline leukaemia virus: Half a century since its discovery

Brian J. Willett *, Margaret J. Hosie

Medical Research Council-University of Glasgow Centre for Virus Research, Institute of Infection, Immunity and Inflammation, College of Medical, Veterinary and Life Sciences, Bearsden Road, Glasgow G61 1QH, Scotland, United Kingdom

* Corresponding author. Tel.: +44 141 330 3274.
E-mail address: Brian.Willett@glasgow.ac.uk (B.J. Willett).

Abstract

In the early 1960s, Professor William F.H. Jarrett was presented with a time-space cluster of cats with lymphoma by a local veterinary practitioner, Harry Pfaff, and carried out experiments to find if the condition might be caused by a virus, similar to lymphomas noted previously in poultry and mice. In 1964, the transmission of lymphoma in cats and the presence of virus-like particles in the induced tumours that bore a striking resemblance to 'the virus of murine leukaemias' was reported in *Nature*. These seminal studies launched the field of feline retrovirology and research on the virus that had been identified and subsequently named 'feline leukaemia virus' (FeLV). This review considers the way in which some of the key early observations made by Professor Jarrett and his coworkers have developed over the years and provides an insight into the substantial progress that has been made since the virus was first discovered.

Keywords: Feline leukaemia virus; Receptors; Pathogenesis; Transmission; Vaccination; Diagnosis

An introduction to feline leukaemia virus

It is thought that the first felid-like carnivores evolved approximately 35 million years ago during the Oligocene epoch. The earliest fossil records of the *Felinae* date from the late Miocene epoch approximately 9 million years ago with remnants of the now extinct *Felis attica* from Eurasia (de Beaumont, 1964); however the domestic cat lineage, the genus *Felis*, appeared closer to 6.2 million years ago during the late Miocene epoch (Johnson et al., 2006). Some time after the emergence of the *Felis*, there appears to have been trans-species infection with gammaretroviruses of rodent origin, giving rise to the ancestral feline leukaemia virus (FeLV).

Nucleic acid sequences related to ancestral FeLVs are present in the genomes of both domestic cats and closely related members of the genus *Felis* with their origins in the Mediterranean basin, including the jungle cat (*F. chaus*), sand cat (*F. margarita*) and European wildcat (*F. sylvestris*), but are absent from other members of the genus from sub-Saharan Africa, Southeast Asia and the Americas, e.g. Geoffroy's cat (*F. geoffroyi*) and the Serval (*F. serval*) (Benveniste et al., 1975). FeLV is now a widespread pathogen of the domestic cat and is distributed worldwide. Befitting its zoonotic origins, the virus has recently crossed genera once again, into the Florida panther (*P. concolor*) and the Iberian Lynx (*L. pardinus*) (Brown et al., 2008; Cunningham et al., 2008; Luaces et al., 2008; Meli et al., 2009, 2010).

Like all retroviruses, FeLV is unstable in the environment and thus transmission is thought to require intimate contact between animals; whether friendly contacts such as grooming, or through fighting and biting (Francis et al., 1977, 1979a and b). A possible exception to the received wisdom is the finding that FeLV may be shed in urine and faeces

and that the shed virus retains the potential for transmission, presenting a theoretical risk of infection (Gomes-Keller et al., 2009).

Following transmission, the course of infection may follow either of two paths. Most commonly, the exposed cat recovers and becomes solidly immune. However, in a lesser proportion of infected cats, the virus establishes a persistent infection that is marked by a viraemia and which is associated with an increased likelihood of developing a severe and ultimately fatal disease. The diseases associated with persistent infection are primarily disorders of haematopoiesis such as lymphomas and leukaemias, immune suppression and degenerative conditions including anaemia. During the prolonged viraemic phase, the animal may present as ostensibly healthy and thus may transmit infection to naïve recipients through virus shed in saliva (Francis et al., 1977; Gomes-Keller et al., 2006). Fortunately, diagnostic tests are available that can detect viraemic cats, facilitating their identification and isolation. Moreover, susceptible cats can be protected from infection by administration of one of the many highly efficacious recombinant FeLV vaccines that are currently available.

Biology and replication of feline leukaemia virus

FeLV belongs to the genus *Gammaretrovirus,* together with retroviruses of rodents, gibbons and koalas. The genetic information of retroviruses is encoded by an RNA genome and the first stage in the viral replicative cycle involves the copying of this RNA genome into a DNA copy. This process is performed by reverse transcriptase, the enzyme that is the target of anti-retroviral drugs such as AZT (azidothymidine, Zidovudine) and d4T (2'-3'-didehydro-2'-3'-dideoxythymidine, Stavudine). Reverse transcriptase subverts the normal cellular flow of genetic information from DNA to RNA to protein. Once a DNA copy of the RNA genome has been synthesised, it integrates into the genome of the target cell as a provirus, assisted by a

second viral enzyme, the ‘integrase’. This provirus remains in the genome of the cell for the lifespan of the cell and, upon cellular division, the provirus is expressed, leading to the production of progeny virions and virus shedding.

It is this unique property of retroviruses that renders them so difficult to eradicate completely from the infected host; a ‘latent’ infection may persist where the virus remains integrated into the genome of cells. Accordingly, both ‘recovered’ cats and solidly immune vaccinated cats may harbour integrated proviral genomes, but may never shed infectious virus; the cats are ‘protected’ from the development of viraemia and thus disease (Torres et al., 2005; Hofmann-Lehmann et al., 2008).

Gammaretroviruses bud from the membrane of the infected cell, undergoing the final phase of maturation with cleavage of the of the Pr65Gag precursor into the mature structural proteins p15 MA, p27 CA and p10 NC. The capsid protein p27 forms the target for the majority of in-practice tests for FeLV, being abundant in the plasma of viraemic cats. However, its utility as a diagnostic for FeLV viraemia is only possible because cats do not appear to respond serologically to the protein, an observation that has led to speculation that cats may be largely immunologically tolerant to p27 through exposure to endogenously expressed FeLV Gag proteins.

As they bud from the infected cell, nascent virions acquire the envelope glycoprotein Env, comprising the surface (SU) glycoprotein gp70 and transmembrane (TM) protein p15E. Gp70 is the target for neutralising antibodies in recovered cats and is an essential component of FeLV vaccines; a recombinant bacterially-expressed form of gp70 is the sole component of one highly effective sub-unit vaccine (Marciani et al., 1991). During the production of this

subunit vaccine, the entire gp70 plus 34 amino acids of the transmembrane domain p15E are expressed in bacteria and purified. As bacterially expressed proteins are not glycosylated, the recombinant protein is referred to as p45.

The combination of p45, the bacterially expressed recombinant Env in aluminium hydroxide and QuilA adjuvant is sufficient to induce protective immunity in vaccinated animals. Immunologically, this would suggest that there is a humoral response directed against linear determinants on gp70 and/or a cellular immune response that confers immunity to infection. While immunised cats mounted a potent humoral response to the vaccine antigen, neutralising antibodies were detected in only a proportion of immunised cats prior to challenge; nevertheless, solid protection was conferred in all animals (Marciani et al., 1991). The absence of neutralising antibodies prior to challenge following vaccination with efficacious FeLV vaccines has been noted for other FeLV vaccine preparations (Hofmann-Lehmann et al., 2007).

Three major subgroups of feline leukaemia virus: A, B and C

In addition to being the primary target for the protective humoral immune response, gp70 also bears the determinants of the viral subgroup. There are three major subgroups of FeLV: A, B and C. The three subgroups were identified by classical assays of viral interference in which prior infection of cells with one subgroup of FeLV prevented subsequent infection with murine sarcoma virus pseudotypes bearing an envelope glycoprotein of the same subgroup (Sarma and Log, 1973, 1977).

Interference assays implied the existence of three distinct viral receptors on target cells. Thus in cells infected with a subgroup A virus, the subgroup A receptor would be either

obscured or down-regulated from the cell surface by interactions with endogenously-produced Env protein. These cells would be rendered refractory to infection with viruses bearing subgroup A Envs, but would remain susceptible to infection with viruses bearing the subgroups B or C Envs.

Using this simple technique, major advances were made in our understanding of the role of the viral subgroups in the pathogenesis of FeLV associated disease. For example, FeLV-A is the most abundant form of the virus in nature and is the virus that is responsible for transmission between animals (Jarrett and Russell 1978; Jarrett et al., 1978). Thus, vaccination against subgroup A virus is the sole requirement for inducing immunity to infection.

The two other subgroups of virus, subgroups B and C, arise in subgroup A infected cats following the establishment of a viraemia. The generation of FeLV-B occurs following recombination between the exogenous FeLV-A and endogenously expressed FeLV Env encoding transcripts (Stewart et al., 1986; Overbaugh et al., 1988). Much has been learned about the recombination breakpoints and the genomic loci responsible for generating subgroup B viruses (Roy-Burman, 1995); the process of recombination between exogenous FeLV-A and endogenous FeLV sequences that occurs in vivo has been reproduced in vitro (Sheets et al., 1992).

The precise site of recombination between FeLV-A and endogenous FeLV in the infected cat is unknown. Previous studies have demonstrated the presence of endogenous FeLV transcripts in the feline placenta (Busch et al., 1983) and in the thymus and lymph

nodes of specific-pathogen free cats (McDougall et al., 1994); thus viral replication at these sites would offer an opportunity for recombination to occur.

As the level of endogenous FeLV transcripts expressed in peripheral blood varies between distinct cat populations and is influenced by breed and gender (Tandon et al., 2007), the likelihood of subgroup B viruses arising in vivo may vary from cat to cat. Early experiments by Bill Jarrett and his colleagues into the association between viral subgroups and disease outcomes demonstrated that cats that were viraemic with FeLV-A and FeLV-B were at a higher risk of developing lymphoma than cats with FeLV-A alone.

With his expertise in animal and human pathology, and his knowledge of neoplastic diseases of haematopoietic and lymphoid tissues (Jarrett and Mackey, 1974), Bill Jarrett observed a predominance of T cell lymphomas in FeLV-infected cats, inspiring the U.S. biomedical researcher Robert Gallo to search for a viral agent associated with human T cell leukaemias. These studies led to the discovery of human T cell leukaemia virus type I and subsequently human immunodeficiency virus type 1.

The third viral subgroup, FeLV-C, arises infrequently in viraemic cats and is only isolated from cats with disease (Jarrett et al., 1978). In a survey of the viral subgroups in naturally-infected viraemic cats, subgroup C viruses were not isolated from 98 clinically normal cats, but were identified in samples from 9/32 anaemic cats surveyed (Onions et al., 1982). The genesis of subgroup C viruses in the infected animal has yet to be ascertained. Molecular analyses have demonstrated that FeLV-Cs have unique receptor binding domain sequences in gp70 that are suggestive of a mutational rather than recombinatorial origin (Riedel et al., 1986; Rigby et al., 1992). FeLV-Cs are not transmitted between cats; rather

they arise de novo in viraemic animals. Following their emergence in vivo, FeLV-Cs give rise to pure red cell aplasia, an anaemia that may prove fatal within weeks.

Cellular receptors for feline leukaemia virus

The virus-receptor interaction is the primary determinant of cell tropism and contributes significantly to the pathogenesis of disease. Accordingly, the cell tropism of the three major subgroups of FeLV is determined by the expression patterns of their respective receptors; FeLV-A Env binds to target cells through an interaction with the thiamine transporter THTR1 (Mendoza et al., 2006), FeLV-B attaches via a phosphate symporter Pit-1 (O'Hara et al., 1990; Johann et al., 1992; Takeuchi et al., 1992) and the closely-related molecule Pit-2 (Anderson et al., 2001), while FeLV-C utilises a haem transporter FLVCR1 for viral entry (Tailor et al., 1999; Quigley et al., 2000) (Fig. 2).

THTR1 is expressed widely in feline tissues, consistent with the broad cellular tropism of FeLV-A (Mendoza et al., 2006). In contrast, although the subgroup C receptor FLVCR1 is expressed widely in haematopoietic tissues (Tailor et al., 1999), its function in the export of cytoplasmic haem would appear to be essential for the development of erythroid progenitors, protecting them from haem toxicity (Quigley et al., 2004). Perturbation of this essential function of FLVCR1 in erythroid progenitor cells results in the selective loss of colony forming units-erythroid, CFU-E, and the development of a severe non-regenerative anaemia.

Thus the seemingly esoteric endeavour of identifying the cellular receptor for FeLV-C has revealed the biochemical basis for the pathological changes described by Hoover et al. (1974) in the bone marrow of cats infected with the KT strain of FeLV (Kawakami et al., 1967) that was subsequently shown to be a mixture of subgroups A, B and C, as well as by

Mackey et al. (1975) following inoculation of cats with the first isolate of FeLV-C. FLVCR1 was the first mammalian haem transporter to be described (Quigley et al., 2004).

The similarities between FeLV-C-associated pure red cell aplasia and Diamond Blackfan anaemia, a rare inherited blood disorder that is characterised by moderate to severe reduction in the production of erythroid progenitor cells, stimulated interest in a role for the human orthologue of FLVCR in Diamond Blackfan anaemia. Subsequent studies revealed that a defect in splicing of the human FLVCR1 gene associated with mutations in the RPS19 gene (Willig et al., 1999; Draptchinskaia et al., 1999) disrupts FLVCR1 expression and contributes to the failure of erythropoeisis in Diamond Blackfan anaemia (Rey et al., 2008), illustrating the value of such comparative studies.

FLVCR1 is not the only receptor for FeLV-C; just as FeLV-B can utilise the closely related Pit-1 and Pit-2 molecules for infection, some strains of FeLV-C can infect through an interaction with FLVCR2 (Shalev et al., 2009). Like FLVCR1, FLVCR2 is a transporter of haem, although its function is in importation of haem (Duffy et al., 2010). Together, the human orthologues of FLVCR1 and FLVCR2 appear to play critical roles in haemostasis. Recently, mutations in FLVCR2 have been found to be associated with Fowler-syndrome (Meyer et al., 2010), a proliferative vascular disorder of the brain.

At present there is no direct evidence to suggest that an impairment of THTR1 function in cats by FeLV-A replication contributes to the diseases that are associated with FeLV-A infection, however it is worthy of note that mutations in the human orthologue of THTR1 are responsible for a thiamine responsive megaloblastic anaemia known as Rogers syndrome (Labay et al., 1999). It is intriguing that one of the earliest studies on the

association between FeLV subgroups and the development of anaemia observed that subgroup A and subgroup C viruses induced distinct anaemias; while FeLV-C induced an aplastic anaemia, infection with a subgroup A virus resulted in a macrocytic anaemia associated with increased erythropoeisis (Mackey et al., 1975).

Transmission and pathogenesis

The original transmission experiments with 'cat leukaemia' were attempted because Bill Jarrett found a cluster of eight cases in a single household of unrelated cats (Jarrett, 1972). Subsequent epidemiological studies by researchers in the U.S. indicated that the introduction of an infected male cat into households for breeding purposes led to the establishment of infection within the household (Brodey et al., 1970), while housing uninfected kittens with infected kittens led to the spread of the virus (Rickard, 1969). Together, these findings suggested to Bill Jarrett that the virus was transmitted horizontally and therefore he conducted a series of experiments which proved that virus transmission occurred between in-contact animals and that physical separation prevented infection, consistent with horizontal transmission (Jarrett, 1972; Jarrett et al., 1973).

The primary route by which FeLV is transmitted is thought to be via oro-nasal exposure to virus-containing secretions. High levels of FeLV are present in the saliva of viraemic cats (Francis et al., 1977, 1979a; Gomes-Keller et al., 2006) but as the virus is relatively labile in the environment (Francis et al., 1979b), it is thought that intimate contact between animals during grooming, sharing feeding bowls or through fighting are the most likely routes of transmission.

The application of highly sensitive molecular techniques to the detection of FeLV RNA in secretions have confirmed the link between the level of plasma viraemia and the amount of virus shed in secretions (Gomes-Keller et al., 2006, 2009); thus a viraemic cat is more likely to transmit virus than a cat that has recovered from infection. Whether a cat recovers from infection or develops viraemia and ultimately develops disease is governed by a number of factors at the time of transmission. One of the key determinants of susceptibility to infection is the age of the cat at the time of exposure (Hoover et al., 1976); only 15% of cats aged between 4 months and 1 year of age became persistently viraemic following experimental infection, while 85% of cats between 2 and 8 weeks of age became infected following challenge (Hoover et al., 1976).

The rapid development of resistance to infection appeared to be immune related, since immunosuppression with either corticosteroids or methylnitrosourea abrogated the age-related resistance (Rojko et al., 1979; Schaller et al., 1978). Recovery from infection is associated with a marked reduction in plasma viraemia, such that virus may no longer be isolated from plasma and virus neutralising antibodies (VNAs) are detected. Recovered cats may also be referred to as having ‘regressive’ infections, while viraemic cats have ‘progressive’ infections.

It is likely that it is the ability of the majority of cats to recover from natural exposure to FeLV that underlies the success of many FeLV vaccines; if 85% of cats between 4 months and 1 year of age recover naturally, then vaccination may simply tip the balance in favour of recovery by priming the immune system to generate robust cellular and/or humoral responses. In support of this hypothesis, most animals vaccinated in an experimental setting do not generate VNAs until after viral challenge, suggesting the induction of an anamnestic response.

A proportion of cats which recover from natural infection with FeLV harbour infectious virus in their bone marrow (Madewell and Jarrett, 1983; Rojko et al., 1982). This state of latency can be reversed by immunosuppressive treatment with corticosteroids (Rojko et al., 1979). The application of sensitive molecular techniques, such as quantitative real-time PCR has provided further insight into the nature of the process of natural recovery from infection, leading to the suggestion that the spectrum of outcomes following infection may require further refinement (Torres et al., 2005; Hofmann-Lehmann et al., 2006, 2007, 2008).

The absence of FeLV proviral DNA after challenge indicates an 'abortive' infection, which is distinct from a 'regressive' infection, where a transient antigenaemia is followed by the establishment of low to moderate proviral loads (Hofmann-Lehmann et al., 2006, 2008). Animals that recovered from infection and which tested negative for viral antigen in plasma, but maintained detectable levels of plasma viral RNA, were considered to be at a higher risk for reactivation of the latent infection than cats which had undetectable plasma viral RNA following recovery (Hofmann-Lehmann et al., 2007). In contrast, viraemic cats were considered to have a progressive infection and this was marked by persistently high plasma viral RNA loads.

Data regarding latently infected cats have been conflicting, while one study reported the presence of detectable proviral DNA and plasma viral RNA (Torres et al., 2005), a second study found no difference in viral and proviral loads between latently infected cats and cats with a regressive infection with no latent infection (Hofmann-Lehmann et al., 2007). Given the discrepant findings, we would support the retention of the definition of a 'latent' infection

as the absence of virus/viral antigen in blood in the presence of reactivatable virus in the bone marrow (Rojko et al., 1982; Madewell and Jarrett, 1983; Hofmann-Lehmann et al., 2008).

Evidence suggests that the latent state is transitory and that, with time, the majority of latently infected cats will ultimately clear the virus from their bone marrow. In a follow-up of the seminal study on viral latency (Madewell and Jarrett, 1983), it was noted that, by 3 years post-infection, only 8% of recovered cats with VNAs still harboured reactivatable proviruses in their bone marrow (Pacitti and Jarrett, 1985). Cats which have recovered from FeLV infection have a similar life expectancy to cats that have never been exposed to FeLV, suggesting that a latent infection with FeLV does not predispose the cat to the development of FeLV-associated disease.

Vaccination against feline leukaemia virus

Ten years after the first description of FeLV, Bill Jarrett published the first reports describing experimental vaccination against FeLV (Jarrett et al., 1974, 1975). This first approach to vaccination relied upon the ability to maintain FeLV-infected cells in culture. Initially, animals were inoculated with either FeLV-infected feline fibroblasts or lymphoblasts and strong humoral responses were induced in either naïve cats or cats previously exposed to FeLV (Jarrett et al., 1974). A refinement of the approach used paraformaldehyde inactivated cells (derived from FL74 lymphosarcoma cells; Theilen et al., 1969) to remove the possibility of an infection being established by the vaccine (Jarrett et al., 1975).

Subsequent studies demonstrated that the proportion of cats challenged with FeLV that became viraemic could be reduced by vaccination with inactivated whole virus derived from FL74 cells (Pedersen et al., 1979), while concentrated envelope glycoprotein purified from the

culture fluid of FL74 cells was sufficient to protect against viraemia, most strikingly when combined with a potent adjuvant (Lewis et al., 1981). By 1985, 10 years from the first experimental inactivated FeLV-infected cell vaccine, the subunit approach to vaccination had been refined, with the incorporation of the viral gp70/85 into immune stimulating complexes (ISCOMs), resulting in the FeLV ISCOM vaccine that protected 100% of cats from viraemia following challenge with FeLV (Osterhaus et al., 1985).

FeLV vaccines have continued to advance over recent years with three main families of vaccine available commercially: (1) classical inactivated virus vaccines, the technology behind which has changed very little over the intervening years; (2) subunit vaccines based on bacterially-produced envelope glycoprotein, gp70 (Marciani et al., 1991); and (3) a live recombinant canary-pox virus engineered to express FeLV genes (Tartaglia et al., 1993; Poulet et al., 2003).

As indicated above, perhaps the most surprising vaccine success has been the bacterially-produced gp70. When Bill Jarrett and colleagues described their first attempts at FeLV vaccination, infected cells were used because it was ‘considered likely that the integrity of the glycoprotein complexes which are effective immunogens might be well preserved’ (Jarrett et al., 1975). This would seem an entirely rational approach to vaccine design. One of the most successful vaccines developed subsequently was the FeLV-ISCOM vaccine which relied on the incorporation of fully glycosylated protein into a complex matrix that ensured delivery of the gp70 to antigen-presenting cells in a conformation that would reflect that on the native virion (Osterhaus et al., 1985).

That a non-glycosylated, bacterially-produced Env protein bearing a C-terminal truncation should protect cats from viraemia (Marciani et al., 1991) must reveal a great deal about the nature of immunity to FeLV. The majority of older cats recover from FeLV infection; vaccination may simply prime cats such that recovery is more likely, if not the sole outcome following challenge. VNAs are seldom induced prior to challenge; however the development of VNAs following challenge is a consistent feature of vaccine induced recovery. The majority of studies suggest that sterilising immunity is seldom induced by vaccination, that a minimal level of viral replication occurs in the vaccinated cat and that this results in proviral integration. However, the vaccinated cat is protected from the development of viraemia and thus will neither shed nor transmit virus, becoming, effectively, a dead-end host for FeLV.

Significant concerns have been raised regarding over-vaccination of companion animals and, in particular, the occurrence of vaccine-associated injection site sarcomas in cats. It is thought that a potent local inflammatory response at the injection site may predispose the animals to the development of soft-tissue sarcomas and thus both strong adjuvants and immunogens have been postulated to play a role.

In order to circumvent the need for strong adjuvants, whilst still inducing robust humoral and cellular immunity, a canary pox (ALVAC) vectored FeLV vaccine has been developed (Tartaglia et al., 1993). ALVAC-FeLV is a replication defective live virus vaccine and thus presents antigen through both the endogenous and exogenous pathways, ensuring the induction of both cellular and humoral immunity. Significantly, ALVAC-FeLV does not require an adjuvant to induce protection from both viraemia and latency (Poulet et al., 2003). Moreover, the large genome of the poxvirus vector allows the incorporation of both *gag* and

env gene products, ensuring a broad antiviral immune response is induced. Given that previous studies have indicated a role for cytotoxic T cells in immunity to FeLV infection (Flynn et al., 2000, 2002), the presentation of T cell epitopes in both Gag and Env from a vectored virus vaccine may prove beneficial in inducing long-lasting immunity and facilitating the efficient clearance of virus-infected cells.

Diagnosing feline leukaemia virus infection

The control of FeLV in a population of cats may be achieved by a combination of isolating infected cats whilst vaccinating susceptible cats; it is recommended that at risk cats be tested prior to vaccination against FeLV. Early studies by Bill Jarrett demonstrated the presence of antibodies in the sera of cats that reacted with feline-oncornavirus-associated cell membrane antigens (FOCMA) and that this reactivity could be used to assess the prevalence of FeLV in a cat population (Jarrett et al., 1973). Accordingly, he was able to show that, in Glasgow, approximately 40% of cats had been exposed to FeLV, whereas the actual incidence of leukaemia in the population was closer to 0.05%, suggesting that the majority of cats exposed to FeLV recover from infection. These studies complemented those in which 'group-specific' or 'type-specific' reagents were used to demonstrate the presence of viral antigens in leukocytes (Hardy et al., 1973).

The majority of in-practice tests for FeLV in current use detect viral antigen in blood, whether whole blood, plasma or serum. The antigenic target for these tests is the viral capsid protein p27, the viral protein that is most abundant in the plasma of viraemic cats. Most in practice tests are based on the principle of rapid immunomigration (immunochromatography). These tests tend to be of high specificity, detecting near to 100% of antigenaemic cats. The sensitivity of these tests is less reliable; a proportion of FeLV-infected cats (regressive

infections) will be antigen-negative and be undetected by antigen-based tests. Detection of these cats relies on more sensitive techniques using PCR and many diagnostic laboratories now offer real-time PCR for the quantification of FeLV proviral DNA and viral RNA.

Quantifying proviral DNA enables the diagnostic laboratory to identify recovered cats (regressive infection) in which the viraemia has been cleared but where proviral DNA may remain integrated into the genome of cells within the bone marrow or lymphoid tissues (Torres et al., 2005; Hofmann-Lehmann et al., 2008). In contrast, real-time PCR for viral RNA is effectively measuring levels of virus in plasma or secretions that are below the sensitivity of tests for viral antigen. Accordingly, it may be possible to detect very low levels of virus in saliva where conventional antigen tests have proved negative (Gomes-Keller et al., 2006). Either test may be of value in ascertaining the biological basis for discordant p27 antigen tests, for example sporadic weak p27 positive tests or borderline positive tests may indicate a latent infection in the bone marrow. Real-time PCR testing may be used to confirm the presence of an infection while quantifying the viral load.

Following a confirmed diagnosis of infection, infected animals should be isolated from susceptible cats to prevent the spread of infection. Testing susceptible cats for evidence of spread will allow the introduction of a vaccination program for the at-risk animals in order to curtail the spread of the virus. Through rigorous testing, vaccination and through the isolation of infected animals, levels of FeLV infection in many parts of Europe have decreased markedly (Lutz et al., 2009).

Conclusions

In 2011, veterinary science lost one of its great investigators. There are many significant scientific breakthroughs in which Bill Jarrett was at the forefront. For us, he will be best remembered for his ground-breaking studies on FeLV. He established a FeLV laboratory which over many years attracted a cohort of scientists who investigated many aspects of the biology of FeLV and established systems that have almost eliminated the virus from the UK.

Conflict of interest statement

None of the authors of this paper has a financial or personal relationship with other people or organisations that could inappropriately influence or bias the content of the paper.

Acknowledgments

When asked to write a review of feline leukaemia virus to commemorate the passing of Professor William F.H. Jarrett, we considered that it would be a fitting tribute to provide an update on some of the more recent developments in our understanding of areas of study that had their origins in the original studies by Bill and his colleagues in Glasgow. Bill was first and foremost a pathologist and ran the hospital pathology service at the Glasgow Veterinary School. His early postgraduate studies with Dan Cappell, Professor of Pathology at the University of Glasgow Medical School, laid the foundation for a career with a broad understanding of both animal and human diseases. Bill had an enquiring mind; he saw patterns in the occurrence of diseases and asked about the causes. Bill was held in very high regard by his colleagues; he was a very exciting person to work with and many of the scientists that passed through his laboratory went on to develop their own subjects and laboratories.

We were fortunate to join the Department of Veterinary Pathology at Glasgow Veterinary School when Bill was the leader of a Department at the forefront of research into AIDS, papillomavirus vaccination and the viral causes of cancer. That companion animal medicine has progressed from the discovery of FeLV to the current day where in many areas FeLV is now on the decline through successful programmes of testing, removal and vaccination is a fitting legacy.

The authors are grateful to Professor Os Jarrett, Bill's brother, for his helpful discussions in the preparation of this review and for his inspiration and mentorship throughout their careers. Work in the authors' laboratory is funded by The Wellcome Trust.

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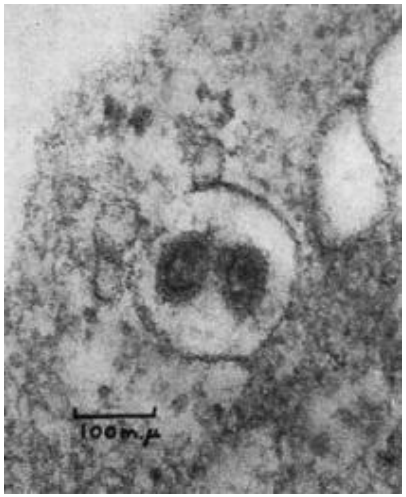
Figure legends

Fig. 1. The assembly of feline leukaemia virus (FeLV) at the plasma membrane. The precursor of the viral structural proteins Pr65Gag is thought to attach to the underside of the membrane via an interaction between N-terminal myristate and membrane phospholipids. Once bound, the Pr65Gag proteins multimerise, triggering the membrane to bend around the forming core. Env proteins associate with the nascent particle through their co-localisation on the membrane until an immature particle is formed. A scission event then takes place releasing the immature virion into the extracellular milieu, at which time the viral protease cleaves Pr65Gag into distinct MA, CA and NC proteins. Inset: Electron micrograph of two FeLV particles in a vesicle within a mesenteric lymph node cell, from Bill Jarrett's seminal study (Jarrett et al., 1964).

Fig. 2. The cellular receptors for feline leukaemia virus (FeLV): (1) FeLV-A; (2) FeLV-B; and (3) FeLV-C. The three viral receptors have different transport functions and thus perturbation of their function by FeLV may result in distinct clinical manifestations.

Figure 1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Mature virions
Immature virions
Env incorporation
Release
Genome packaging and budding
Multimerisation
Membrane binding
Targeting
Pr65Gag
Env
Pr65 Gag
gp70
p15E
m
p15 MA
pp12
p27 CA
p10 NC

Figure 2