Figure 1.2. Veterinary Diagnostics Market, by Product (2013-2018)

Source: Veterinary Diagnostics Market by Products (Clinical Chemistry, Hematology Analyzers, Molecular Diagnostics, Immunodiagnostics, Diagnostics Imaging), by Animals (Companion, Food-producing, Dog, Livestock, Poultry, Swine) - Global Forecast to 2018

To 2018, North America will continue to drive and dominate the veterinary diagnostics market. However, the market share of this region will decrease slightly during 2013 to 2018, primarily due to the increase in demand for veterinary diagnostic tests in the emerging economies of Asia-Pacific and Latin America. Most of the growth in the Asia-Pacific region is driven by the increasing demand for diagnostic tests in the animal health industries of China and India. The growth in the Asia-Pacific and Latin American regions can also be partly attributed to the increasing disposable incomes leading to growth in the per capita animal health expenditure in these regions.

The companion animal diagnostics market includes in vitro diagnostic (IVD) products – instruments, consumables, test reagents and test kits – for the diagnosis of disease and health conditions in household pets (primarily dogs and cats) as well as horses. Market performance is determined by diagnostic product demand from veterinary clinics, hospitals, reference labs, and governmental and research laboratories such as those found at universities and associated with government agricultural and animal health authorities.

In-vitro diagnostic (IVD) instrumentation and products have been extensively applied to veterinary healthcare; various assay forms, analyzes and analyzers are transferrable from the clinical space to the veterinary clinic and laboratory. Major diagnostic methods in mainstream human healthcare, immunodiagnostics, molecular techniques (including nucleic acid testing), haematology and clinical chemistry, all now have standard applications in the veterinary care of companion (pets) and food animals (livestock and production animals).

The food animal diagnostics market includes primarily immunodiagnostic test kits, molecular test kits, reagents, other consumables, instruments and other supplies for disease diagnosis in livestock and production animals, commonly cattle, pigs, poultry, sheep and goats. The performance of the food animal diagnostics market is largely determined by the prevalence of infectious food animal diseases in a country or global region, or at least the economic with which this is considered. The penetration of easy-to-use molecular diagnostic test kits for veterinary infectious diseases also represents another factor improving the value of the global food animal diagnostics market.
Molecular Diagnostics Market by Application (Infectious Disease, Oncology, Genetics, Microbiology), Technology (PCR, INAAT, DNA Sequencing), End User (Hospital, Laboratories), Product (Instruments, Reagent, Service, Software) - Global Forecasts to 2018

Diagnosis and disease monitoring in veterinary medicine, particularly for companion animals, has tended to follow trends in human medicine. Being able to track new diseases as they emerge, and making sure we are prepared for them, is essential in order to help veterinary professionals, governments and farming communities to reduce the risk of disease outbreaks and pandemics.

1.3 Factors driving growth in the veterinary diagnostics for infectious diseases market

1.3.1 Global trends in animal production and meat consumption

Over the foreseeable future there is likely to be considerable global growth in animal production. In market terms this will be because there are economic drivers on both the supply and demand side of the equation.

Figure 1.4. The forecast of global demand for meat, 2005 versus 2050

There is general recognition that there will have to be significant increases in production to meet growing demand for livestock products in the coming decades. It has been claimed that meat production will have to double by 2050 to meet growing demand (FAO, 2006). In the same report the FAO suggest that there will be much more growth in production from developing countries than from the developed world (4 million tonnes per year vs 0.8 million tonnes).
is required to realise the potential with well-characterised, validated assay systems that provide improved diagnostic capabilities.

Outbreaks of major animal diseases can have a devastating impact on animal and human health, food safety, the wider economy, the countryside, animal welfare, rural communities and the environment. The use of robust and fit for purpose diagnostic tests are a key component in the control of diseases, having the potential to effectively support control and if appropriate, eradication.

The choice of assay used in a given situation and purpose must be given due attention, consideration and evaluated in the first instance for cost, speed, consistency and accuracy. There is no perfect test; selecting the best test is a question of considering how multiple factors inter-relate to answer specific questions being asked.

Table 2.1. Some important criteria for consideration in selection of an appropriate assay technique.

<table>
<thead>
<tr>
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<th>Criterion</th>
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<tbody>
<tr>
<td>1</td>
<td>What specific question is the assay attempting to answer?</td>
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<td></td>
<td>For example is the animal infected now or has it been in the past?</td>
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<tr>
<td>2</td>
<td>What degree of sensitivity/specificity is needed?</td>
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<tr>
<td>3</td>
<td>Is a rapid result required?</td>
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<tr>
<td>4</td>
<td>Is high throughput required?</td>
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<tr>
<td>5</td>
<td>Are there cost constraints?</td>
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<tr>
<td>6</td>
<td>Is there a suitable assay readily available?</td>
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<tr>
<td>7</td>
<td>Does the assay require specific instruments/equipment?</td>
</tr>
<tr>
<td>8</td>
<td>What levels of operator competency and training are required?</td>
</tr>
<tr>
<td>9</td>
<td>Are there specific regulatory, OIE, National requirements?</td>
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The *OIE Manual of Diagnostic Tests and Vaccines for Terrestrial Animals* specifies and details the internationally agreed diagnostic test methods for all the OIE listed diseases as well as several other diseases of importance.

**Samples for diagnostic assays**

Sample collection, transport, storage and preparation procedures are critical in the diagnostic process. If these following points are not given sufficient consideration, even the most robust of diagnostic methods will fail to produce reliable results.

- Collection
Micro- and nano- particle labels. In diagnostic applications, micro- and nano- particles can be used as labels for selective and specific detection in immunoassays.

Label-less immunoassays. While some kind of label is generally employed in immunoassays, there are certain kinds of assays which do not rely on labels, but instead employ detection methods that do not require the modification or labeling of the components of the assay. Surface plasmon resonance (http://en.wikipedia.org/wiki/Surface_plasmon_resonance) is an example of a technique that can detect binding between an unlabeled antibody and antigens. Another demonstrated labelless immunoassay involves measuring the change in resistance on an electrode as antigen binds to it.

2.3.1.3 Immunoassay configurations

In the early 1900s, it was observed that immune sera from animals immunised with a bacteria would bind firmly and specifically to the same bacteria in vitro (Bordet and Genjou, 1906). This observation was the first qualitative immunochemical characterisation. In the following two decades there were several reported observations that the serum from an immunised animal would render a bacterial broth culture turbid. During the subsequent years it was realised that the reaction of antibody and antigen produces a precipitate in a gel matrix. The precipitate could then be used to characterise the reagents and under certain conditions could be used to quantify the reagents. This gave rise to the gel immunodiffusion techniques that have been widely used in diagnostic laboratories from the 1940’s through to the current day.

2.3.2 Agglutination assays

Agglutination assays were first introduced in the 1920’s using sheep erythrocytes. Later in the 1950’s, the first assays using latex particles were described (Wild, 2013). The principle is that antibodies bind to the surface of the particles or blood cells, which form aggregates when a polyvalent antigen is present in a sample. Alternatively the particles may be first coated with antigen, for the detection of antibody. Agglutination inhibition assays (e.g. haemagglutination inhibition assay, HI) have been widely used e.g. in influenza. In this method, a specific antigen is added to the sample, if complementary antibody is present, it will bind the antigen and inhibit antibody induced agglutination of antigen coated particles. In either format, the results may be read by eye or with the aid of a microscope. A quantitative result can be obtained by the titration of antigen (or antibody) in a dilution series and the data compared to a reference standard.
In Spain animal disease diagnostic tests are regulated by the Ministry of Agriculture, Food and Environment. A technical dossier has to be submitted by the manufacturing company.

### 3.5.2 Regulation of Veterinary Diagnostics in USA

In the USA, APHIS regulates veterinary biologics (vaccines, bacterins, antisera, diagnostic kits, and other products of biological origin), this work is done by APHIS’ Centre for Veterinary Biologics (CVB) and is centered around enforcement of the Virus Serum Toxin Act.

The CVB regulates test kits, defined as all critical reagents needed for testing, packaged together with instructions for use and interpretation of results, but does not regulate individual reagents. According to the CVB criteria, a test kit must claim to detect or diagnose the existence of, or susceptibility to, potential disease causing agents in animals. Test kits which do not claim to detect or diagnose an infectious disease (e.g. bacterial typing or metabolic disease kits) are not regulated by the CVB.

The CVB require submission of the following data to support application for a licence:

- Diagnostic sensitivity/specificity studies – comparing the test to an existing gold standard test (as specified by CVB).

- Reproducibility/repeatability/suitability studies – using a blinded panel of sera.

- Ruggedness/robustness studies – measures the capacity of the assay to remain unaffected by deliberate small variations in method parameters.

All label claims must be specific and supported by data filed with the CVB. For diagnostic test kits, labelling should specify:
of laboratory evaluation. Recently, methods for the detection of parasitic antigens or nucleic acids have made considerable progress.

### 4.2.1 Basic diagnostic parasitological techniques

The traditional techniques for the diagnosis of intestinal parasites (helminth and intestinal protozoa) are the formol-ether concentration method (FECM) or direct faecal smear. For quantitative diagnosis of soil transmitted helminths the Kato-Katz method is often used. Faecal egg count techniques for example FLOTAC and McMaster are also in common use.

http://journals.plos.org/plosntds/article?id=10.1371/journal.pntd.0001201#pntd-0001201-g005

The FLOTAC ® method was developed by Professor Giuseppe Cringoli (University of Naples, Italy) to detect and quantify the number of eggs, oocysts, cysts and parasite larvae found in animals and humans.

**Figure 4.1. Illustration of the principle of the Flotac method**

http://www.dolf.wustl.edu/?page_id=1302
The variability of the clinical signs and post-mortem lesions mean that these do not provide firm evidence for the unequivocal diagnosis of CSF. There are a number of viral and bacterial diseases that may be confused with acute CSF, e.g. African swine fever (ASF), porcine dermatitis and nephropathy syndrome (PDNS), post-weaning multisystemic wasting syndrome (PMWS), thrombocytopenic purpura, salmonellosis, erysipelas, pasteurellosis, actinobacillosis and Haemophilus parasuis. A tentative diagnosis based on clinical signs and post-mortem lesions must therefore be confirmed by laboratory investigation. Laboratory methods for diagnosis of CSF are aimed at detection of the virus, viral nucleic acid, viral antigens or detection of specific antibodies. Isolation in cell culture, from samples of tonsil is considered the most sensitive method for virus isolation. Alternatively spleen, kidney, ileum or lymph nodes can also be used.

Figure 5.1. Characteristic pinpoint haemorrhages on sliced kidney surfaces in classical swine fever

RT-PCR methods have been described for the detection of CSF in infected animals during the early incubation period and for a longer period of time in cases where the pigs recover. Assays are available for both CSFV antigen and antibody detection. Typically samples collected for antigen detection are peripheral blood leucocytes, whole blood and tissues. Serum and plasma are collected for the detection of antibody. ELISA’s using polyclonal and monoclonal antibodies, together with either native or recombinant antigens are available. These tests can be used to differentiate vaccinated from infected animals if directed against the ‘Erns’ protein if used in conjunction with the ‘E2’ marker vaccine, of which there are several commercial products. Commercial assays for the Erns protein are available from IDEXX, Synbiotics and Prionics.

5.3 Bovine Herpes Virus (BHV)

BHVs are known to cause a variety of syndromes in cattle, most notably infectious bovine rhinotracheitis (IBR). In addition to causing respiratory disease, bovine herpesvirus 1 (BHV-1) can cause conjunctivitis, vulvovaginitis, abortions, encephalitis and generalised systemic infections (Wyler et al., 1989). Clinical findings may be highly suggestive of a diagnosis of IBR but a definitive diagnosis must be made in the laboratory. The virus can be detected by viral isolation on specimens collected during the febrile acute phase of the infection. BHV-1 can be readily isolated in cell culture from nasal swabs, conjunctival swabs, vaginal swabs, preputial washings, placental cotyledons of aborted foetus, foetal liver, lung, spleen, kidney, lymph node, mucous membrane of respiratory tract, tonsils and lungs collected in virus transport medium.
6.4 ZOETIS

http://www.zoetis.com

Zoetis formerly Pfizer Animal Health entered the veterinary diagnostics market by the acquisition of Synbiotics. This was seen as an opportunity to offer a range of diagnostic tests that would broadly complement the Zoetis portfolio of animal health products and services, thus providing continuity through from disease detection and diagnosis to treatment, prevention and control.

In December 2010, Pfizer Animal Health (now Zoetis) announced an agreement to acquire Synbiotics Corporation (PinkSheets SYNB.PK), a privately held, Kansas City-based leader in the development, manufacture and marketing of immunodiagnostic tests for companion and food production animals.


Synbiotics was a leading developer and manufacturer of veterinary diagnostics and services for veterinary practices, livestock and poultry producers, reference laboratories and animal breeders worldwide. New product R&D was centered at the Synbiotics headquarters in Kansas City, Missouri and also performed in manufacturing locations in Lyon, France and in San Diego, California, and through R&D collaborations with the University of Maryland and Kansas State University.
Assays for other species

LifeAssays® VetReader System is a commercialised system for point-of-care testing of canine CRP and feline and equine haptoglobin. The test utilises a patented magnet immunoassay (MIA) technique.

**Figure 8.1. Diagrammatic illustration of the magnet immunoassay technique**

![Diagram of magnet immunoassay technique](www.lifeassays.com)

Source: www.lifeassays.com

Life Technologies have commercialised the Acute Phase 4-Plex Panel Luminex® platform for quantifying human β2 Microglobulin, Haptoglobin, CRP, and Gc Globulin in serum, citrate plasma, heparin plasma, cerebrospinal fluid, urine, and tissue culture supernatant. The system measures 4 analytes simultaneously.

www.lifetechnologies.com

Avacta Animal Health currently offer testing for APPs in dogs, cats, and horses:

<table>
<thead>
<tr>
<th>Species</th>
<th>APPs</th>
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</table>
| Canine  | C-Reactive Protein  
          | Haptoglobin  
          | Serum Amyloid A (SAA)  
          | Alpha 1 Acid Glycoprotein (AGP) |
| Feline  | Alpha 1 Acid Glycoprotein (AGP)  
          | Haptoglobin  
          | Serum Amyloid A |
| Equine  | Serum Amyloid A |

www.avactaanimalhealth.com

**Accuplex Diagnostics.** An Irish based company started in County Kildare, Ireland. The company specializes in Equichek™ a lateral flow point-of-care diagnostic for the acute phase protein amyloid in performance horses, a marker of infection.

http://accuplexdiagnostics.com/