36 Chapter

Orthomyxoviruses and Paramyxoviruses

ORTHOMYXOVIRUSES

Orthomyxoviruses are so named because of their affinity for mucins. They have three genera – *Influenzavirus A*, *B* and *C*.

Morphology

Influenzavirus A and *B* are morphologically similar, but *Influenza virus C* differs from them in certain aspects, particularly in having **only a single type of glycoprotein spike**. The virions are spherical, 80–120 nm in diameter, but larger pleomorphic and filamentous forms 1,000 nm or more in length may be abundant. The nucleocapsid (Fig. 36.1) has helical symmetry with a core of eight segments of single-stranded RNA of negative polarity and molecular weight of 5,000 kDa. The nucleocapsid is surrounded by a protein layer or **matrix protein M1**, which in turn is enclosed in host plasma membrane derived **lipid bilayer envelope**. The **M2 protein** projects through the envelope to form ion channels which allow pH changes in the endosome.

Attached to the lipid envelope of *Influenzavirus A* and *B* are two types of glycoprotein peplomers or spikes, the haemagglutinin (HA) and the neuraminidase (NA). HA peplomers exist as 500 tapered projections, 10 nm in length, with their broadest (5 nm) ends outermost and their narrow ends inserted in the lipid membrane. NA peplomers are 100 mushroom-shaped projections per virion measuring 9 nm in length. The virus particles bind to the host cell receptors (sialic acid) by haemagglutinin peplomers.

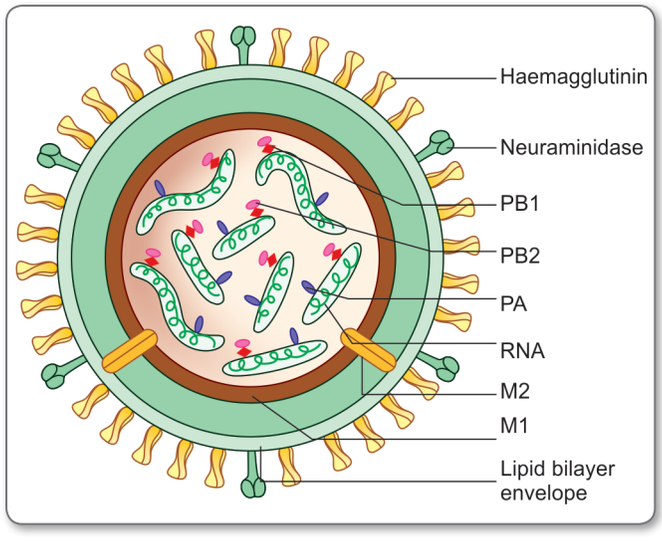


Fig. 36.1: Influenza virus.

Antigenic Variation

Influenza virus is capable of undergoing antigenic variation due to frequent changes in the antigenicity of HA and NA. This is of great importance in the epidemiology of the disease.

Antigenic Drift

The gradual sequential change in the antigenic structure occurring regularly at frequent intervals is known as antigenic drift. It results from mutations in *HA* or *NA* genes. The new antigens acquired though different from the previous are yet related to them so that they can still react with antisera to the predecessor virus strain to some extent. Periodic epidemics of influenza are associated with antigenic drift.

Antigenic Shift

It is an abrupt, drastic, discontinuous variation in the antigenic structure resulting in a new virus strain unrelated antigenically to predecessor strain. It may involve HA or NA or both. Antibodies to predecessor virus strains do not neutralize the new variants, therefore, they can spread widely in the population leading to major epidemics and pandemics.

Pathogenesis

The route of entry of the virus is the respiratory tract. Very small doses, three viable particles or more can initiate infection. The viral neuraminidase facilitates infection by hydrolysing the mucus film lining the respiratory tract and exposing the cell surface receptors for virus adsorption. Ciliated epithelial cells of respiratory tract are the main sites of virus infection. The death and sloughing of these cells renders the respiratory tract vulnerable to secondary bacterial infection. Viraemic phase is extremely rare.

Incubation period is short varying from 1–4 days. The illness is characterized by a sudden onset of systemic symptoms such as

chills, fever, sore throat, nonproductive cough, myalgia, headache and malaise. The uncomplicated illness usually lasts for 3–7 days.

Laboratory Diagnosis

- **Demonstration of virus antigens**
  Smears of nasopharyngeal secretions and nasal swab or centrifuged deposit of throat garglings are stained with fluorescein-tagged influenza antiserum and seen under fluorescence microscope.
- **Virus isolation**
  Influenza viruses can be isolated during first 2–3 days of illness. Throat garglings are collected using suitable buffered salt solution. The specimen should be processed immediately. If short delay is expected then store it at 4°C and if long delay is expected then store it at –70°C. The specimen is treated with antibiotics to destroy bacteria and inoculated into amniotic cavity of chick embryos, primary monkey kidney or human embryo kidney cells or Madin-Darby canine kidney (MDCK) cells.
- **Serological tests**
  Serological diagnosis of influenza is based upon demonstrating a four-fold or higher increase in antibody titre between acute-phase and convalescent-phase sera. Complement fixation, haemagglutination inhibition and neutralization tests are done for this purpose. Since serological diagnosis is retrospective, therefore, it is of limited use.

Prophylaxis

Vaccination is the main method of preventing influenza, but major difficulty in immunoprophylaxis is the frequent change in the antigenic structure of influenza virus. Vaccines are of two types:

- **Inactivated vaccines**
  Influenza virus is grown in allantoic cavity of chick embryos. These are inactivated with formalin or β-propiolactone, then purified by zonal ultracentrifugation and disrupted with detergents. The resulting polyvalent inactivated vaccine is used every autumn. This vaccine can be further purified to contain only haemagglutinins and neuraminidases. This is known as subunit vaccine. Inactivated vaccines induce the formation of circulating antibodies. The level of antibodies in the respiratory mucosa is only a fraction of the serum level.
- **Live attenuated vaccines**
  Temperature-sensitive (ts) mutants of influenza virus may be used as live vaccine. They can grow at lower temperature of the nasopharyngeal mucosa (32–34°C) but not in lungs (37°C). Live vaccine is administered by aerosol spray or intranasally. It stimulates the production of local IgA antibodies. Circulating antibodies of IgM and IgG classes are not raised to the same extent as when killed vaccine is injected.

PARAMYXOVIRUSES

Morphology

Paramyxoviruses include parainfluenza, mumps and measles viruses. They are indistinguishable in the electron microscope. They range from 150–300 nm or more in diameter, occasionally there are filamentous forms and giant forms up to 800 nm in diameter. They are enclosed by a lipid envelope derived from plasma membrane of the host cell. It contains HN and F glycoprotein peplomers 12–14 nm long and 2–4 nm wide. The former carries both haemagglutinating and neuraminidase functions and latter causes fusion of cell membranes leading to the formation of syncytia. The spikes on the meales virus envelope carry a haemagglutinin but not neuraminidase. The inner surface of the envelope is lined by matrix (M) protein. Within the virion, there is a nucleocapsid of helical symmetry. It contains a single-stranded, negative sense RNA genome as a single piece (Fig. 36.2).

PARAINFLUENZA VIRUSES

There are four types of parainfluenza viruses, types 1, 2, 3 and 4.

Pathogenesis

- Parainfluenza viruses are highly transmissible and are acquired by droplets and by contact with respiratory secretions. **Incubation period varies from 2–6 days**. All parainfluenza viruses produce upper respiratory tract infections. In infants and children, these viruses may invade the lower respiratory tract, causing **pneumonia**. In older children (6 months to 5 years), parainfluenza virus type 1 and to some extent, type 2 may cause **laryngotracheobronchitis or croup**. The patient presents with fever, cough and respiratory distress that may need emergency tracheostomy. Type 3 infects infants within first year or two of life. Type 4 produces only mild illness.

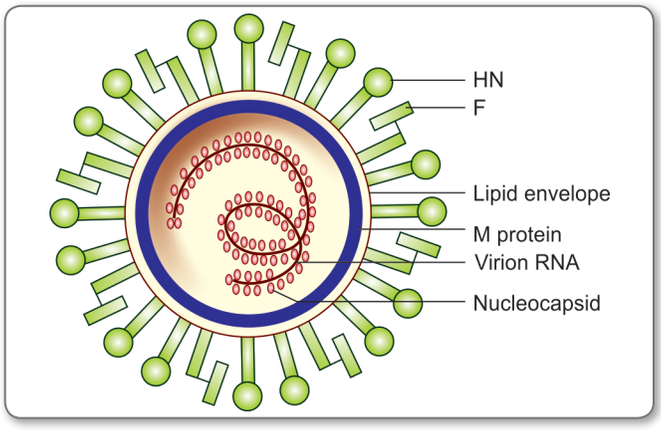
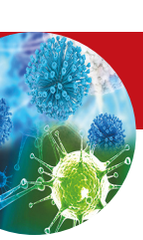


Fig. 36.2: Paramyxovirus.



Laboratory Diagnosis

- Immunofluorescent staining of exfoliated cells aspirated from respiratory tract.
- Detection of free antigen in mucus by ELISA and RIA.
- Parainfluenza viruses may be isolated in primary human or monkey kidney cells or in continuous cell lines such as H292 derived from human lung mucoepidermoid carcinoma. They produce little cytopathic effect, except type 2 which induces syncytia. Viral growth can be detected by haemadsorption of guinea pig red cells or by use of specific immunofluorescent antibody.
- Type-specific antibody may be detected by haemadsorption inhibition, neutralization, ELISA and complement fixation test. A four-fold rise in titre is indicative of infection with a parainfluenza virus.

MUMPS

Pathogenesis

Mumps is predominantly a disease of childhood. The mumps virus is transmitted by way of respiratory and oral secretions and respiratory tract is the portal of entry. It multiplies in the upper respiratory tract and in local lymph nodes. The virus then enters the blood stream and the infection spreads to many organs of the body. The major manifestation is painful swelling of one or both parotid glands occurring 14–18 days after exposure. It may also cause meningoencephalitis, orchitis, oophoritis, pancreatitis, arthritis, myocarditis and renal dysfunction.

Laboratory Diagnosis

- Virus can be isolated from saliva from affected gland, throat swab, CSF and urine on primary monkey kidney cells, H292 and HEp-2 cells. It produces little cytopathic effect but virus can be detected by immunofluorescence and haem-adsorption (of guinea pig or chicken red cells) which can be inhibited by specific antiserum.
- For rapid diagnosis, ELISA is useful for detecting mumps-specific IgM antibodies in the serum.

Prophylaxis

A live attenuated vaccine, derived by passage in chick fibroblasts, offers 95% protection which lasts for 12 years. It can be given by subcutaneous injection in combination with attenuated measles and rubella strains (MMR vaccine). This vaccine is administered to children of both sexes, aged 15 months.

MEASLES

Pathogenesis

Measles is the **commonest highly contagious childhood disease** spread by respiratory secretions. **After an incubation period of 10–12 days**, patient develops upper respiratory tract infection with high fever, rhinitis, cough and conjunctivitis. **Koplik's spots**, which are small, 1–3 mm in diameter, bluish white spots surrounded by erythema can be seen on the buccal mucosa during this stage and are pathognomonic of measles. After 1–2 days, the acute symptoms decline with the appearance of characteristic **maculopapular rash** which appears first on the neck and then spreads to the rest of the body. **The rash is due to type IV hypersensitivity to viral antigens**. In next 10–14 days, rash fades with considerable desquamation and sometimes by discolouration of the skin. There is only one antigenic type of measles virus. Infection confers lifelong immunity.

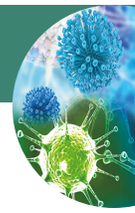
Since measles decreases the resistance of the respiratory epithelium, therefore, patient may develop secondary bacterial infections like sinusitis, otitis media, bronchitis, bronchiolitis, croup and bronchopneumonia. Giant cell pneumonia may occur most frequently in those with impaired cell-mediated immunity. In addition, acute postinfectious encephalitis and subacute sclerosing panencephalitis (SSPE) may occur in 1 in 1000 and 1 in 300,000 cases of measles respectively.

Laboratory Diagnosis

- The measles virus can be isolated, though with some difficulty, from throat washings, blood, urinary sediment, etc. on monkey kidney, primary human embryo kidney and human amnion cells. The appearance of multinucleated giant cells containing numerous acidophilic inclusions in cytoplasm and nuclei, in the cultured cells, suggests the presence of measles virus. For rapid diagnosis, both multinucleated giant cells and viral antigens can be detected in the cells aspirated from nasopharynx.
- Measles antibody in the patient serum can be detected by IgM capture ELISA. Complement fixation test can be carried out on acute and convalescent sera. A rise in antibody titre is diagnostic.

Prophylaxis

Children of the age of 15 months are given MMR vaccine, followed by a booster at the age of 4–6 years.



Key Points

- The nucleocapsid of influenza virus has **helical symmetry** with a core of **eight segments of single-stranded RNA** of negative polarity.
- Attached to the lipid envelope of *Influenzavirus A* and *Influenzavirus B* are two types of glycoprotein peplomers, the **haemagglutinin** (HA) and the **neuraminidase** (NA).
- Influenza virus is capable of undergoing **antigenic variation** by *antigenic drift* and *antigenic shift*.
- ***Mumps virus*** *is the major agent of* ***parotitis***.
- *Measles is an acute febrile infection with an* ***exanthematous rash****; prodromal symptoms of measles include* ***Koplik's spots on the buccal mucosa***.
- **MMR** vaccine prevents **measles**, **mumps** and **rubella**.

Assess Yourself

Long and Short Answer Questions

1. Discuss the morphology and pathogenesis of influenza virus infection.
2. Write short notes on:
   (a) Measles virus
   (b) Mumps virus

Multiple Choice Questions

1. Which of the following genera is **not** included in family Orthomyxoviridae?
   (a) *Influenzavirus A*
   (b) *Influenzavirus B*
   (c) *Influenzavirus C*
   (d) *Parainfluenzavirus*
2. Measles virus belongs to which of the following genera?
   (a) Rubulavirus
   (b) Morbillivirus
   (c) Pneumovirus
   (d) Paramyxovirus
3. Koplik's spots are characteristic of which of the following infections?
   (a) Mumps
   (b) Measles
   (c) Herpes
   (d) Rubella

Answers to MCQs

1. d **2.** b **3.** b

42 Chapter

Medically Important Arthropods

Arthropods belong to the phylum Arthropoda. They are animals with jointed legs, segmented bodies and chitinous exoskeletons. There are over one million species in the phylum Arthropoda (insects and their allies). *Arthron* in Greek means jointed and *poda* means feet. Arthropods are one of the most important sources, other than man himself, of human pathogens. Scientific study of insects is known as *entomology*. *Entom* in Greek means insect and *logas* means scientific study.

Medical entomology is the branch of this science that deals with the arthropods, which are involved with the spread of disease in human beings. Medical significance of arthropods is attributed mainly due to their blood sucking habits, and their role as vectors of the agents of bacterial, viral or parasitic infection. They can transmit infection in the following ways:

- **Mechanical transmission**, in which the arthropod vector merely transports the organisms from one host to another or from the environment to a host and is not a part of the life cycle of the organism.
- **Biological transmission**, requiring a period of incubation or development in this host. For example, malaria parasites undergo multiplication in the female anopheles mosquito before becoming infective for man.

There are five main classes in the phylum Arthropoda (Table 42.1). Class Insecta, class Arachnida and class Crustacea are of medical importance. Other classes include primarily parasites of the animals. Class Insecta includes mosquitoes, flies, fleas, bugs and lice; and class Arachnida includes ticks, mites, spiders and scorpions. Class Crustacea includes cyclops, crabs and crayfish.

Arthropods of medical interest ingest the pathogenic organisms and convey these to man by following methods:

- Arthropods may obtain the parasitic organisms in a blood meal from an infected person and deposit them in a vomit-drop in the puncture wound (plague) or in faecal pellets near the puncture wound (epidemic and endemic typhus, trench fever, and Chagas' disease) made in the skin of an uninfected person.
- Contamination from infected hemolymph may occur when the arthropod is crushed on the skin (epidemic typhus and epidemic relapsing fever).

TABLE 42.1: Classification of medically important arthropods

Class	Order	Common names
Insecta	Diptera	Mosquitoes, sandflies, tsetse flies, blackflies, deerflies, botflies, warble flies, house flies, flesh flies, blowflies and kads
	Hemiptera	Bed bugs and assassin bug
	Coleoptera	Beetles
	Siphonaptera	Fleas
	Anoplura	Sucking lice
	Mallophaga	Biting lice
	Hymenoptera	Wasps, honeybees and ants
	Dictyoptera	Cockroaches
Arachnida	Acari	Hard ticks, soft ticks, chiggers, itch mites and follicle mites
	Araneae	Spider
	Scorpiones	Scorpions
Crustacea	Copepoda	Cyclops
	Decapoda	Crabs and crayfish
Pentastomida		Tongue worm
Myriapoda	Diplopoda	Millipedes
	Chilopoda	Centipedes

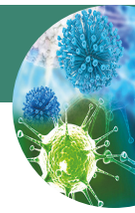


TABLE 42.2: Common arthropods and diseases transmitted by them

Arthropod	Diseases transmitted
Mosquitoes	
• Anopheles	Malaria, filariasis (not in India)
• Culex	Filariasis, Japanese encephalitis, West Nile fever
• Aedes	Yellow fever, dengue, chikungunya haemorrhagic fever, Rift Valley fever, filariasis
• Mansonia	Brugian filariasis
Flies	
• House fly	Typhoid and paratyphoid fever, diarrhoea, dysentery, cholera, gastroenteritis, amoebiasis
• Sand fly	Kala-azar, oriental sore, espundia, oroya fever
• Tsetse fly	African trypanosomiasis
Louse	Epidemic typhus, relapsing fever, trench fever
Rat flea	Bubonic plague, endemic typhus, *Hymenolepis diminuta* infection
Black fly	Onchocerciasis
Reduviid bug	Chagas' disease
Ticks	
• Hard ticks	Spotted fever group, viral encephalitis, Colorado tick typhus, tularaemia, human babesiosis
• Soft ticks	Q fever, relapsing fever, Kyasanur Forest disease
Mites	
• Trombiculid mite	Scrub typhus
• Gamasid mite	Rickettsial pox
• Itch mite	Scabies
Cyclops	Dracunculosis, diphyllobothriasis
Cockroaches	Same as in case of house fly

- Some arthropods discharge the pathogens (malaria sporozoites and virus particles) through the hypopharynx in minute droplets of salivary secretion at the time they procure a blood meal. Filarial larvae from the proboscis (labium and mouth area) of the mosquito are deposited on the skin near the site of the puncture. They then enter through puncture wound or penetrate through the skin on their own.

In many arthropod-associated diseases, the etiologic agents were undoubtedly parasites of their invertebrate hosts long before man came into the life cycle. In some cases the parasite has been so long and so well adjusted to the arthropod that it produces no obvious injury, namely, *Rickettsia rickettsii* and *Borrelia duttoni* in ticks, and *R. tsutsugamushi* in trombiculid mites. In the tick and in the mite the respective parasites are even transmitted vertically (congenitally) and require no vertebrate host for at least several generations. On the other hand, *R. prowazekii* in the body louse can cause extensive and often fatal damage to this ectoparasite, suggesting that the vector-pathogen relationship is an imperfect one. Vector-borne human infections are given in Table 42.2, and biting characteristics of medically important arthropods are given in Table 42.3.

TABLE 42.3: Biting characteristics of medically important arthropods

Only females bite
• Mosquitoes of genera *Anopheles* and *Culex*
• Sandflies of genera *Phlebotomus* and *Lutzomyia*
• Blackflies (*Simulium* spp.)
• Deerflies or mango flies (*Crysops* spp.)
• Biting midges (*Culicoides* spp.)
Both males and females bite
• Tsetse flies (*Glossina* spp.)
Usually bite at night
• *Anopheles* mosquitoes
• Sandflies of the genera *Phlebotomus* and *Lutzomyia*
• Triatomine bugs
Usually bite during the day time
• *Aedes* mosquitoes
• Blackflies (*Simulium* spp.)
• Biting midges (*Culicoides* spp.)
• Deerflies or mango flies (*Chryosops* spp.)
Insects those are able to pass through ordinary mosquito net
• Sandflies of the genera *Phlebotomus* and *Lutzomyia*

INSECTS

Mosquitoes

Mosquitoes are readily recognised by a long needle-like proboscis. Adult males and females both feed on plant juices, but the female needs blood for the development of her eggs, and is also a voracious predator on a wide variety of vertebrate animals throughout the world. Mosquitoes of importance in human medicine are divided into two broad types:

- **Anopheline mosquitoes**, numerous species of which transmit malaria.
- **Culicine mosquitoes**, which are the vectors of many arbovirus infections.

Both anopheline and culicine mosquitoes also act as the intermediate hosts of certain filarial worms. Female mosquitoes lay their eggs on water; larvae and pupae are both aquatic. Most anopheline mosquitoes prefer relatively large expanses of water that do not dry up, but many culicine mosquitoes, particularly *Aedes* spp. will breed in small pockets of water, such as tree holes, water butts, etc. Adults have wide flight range and may be found several kilometres from their breeding ground.

Sandflies

Sandflies are tiny flies that are well able to penetrate ordinary mosquito net. They have a restricted flight range, so that the diseases they transmit – notably kala-azar and other forms of leishmaniasis, bartonellosis and sandfly fever – tend to be localized in distribution. Female flies suck blood, usually at night, and breed in dark, moist areas, often in or around human dwellings. Species associated with disease transmission in Africa, the Middle East, Asia and the Mediterranean littoral belong to the genus *Phlebotomous*. In Central and South America, *Lutzomyia* spp. act as vectors of leishmaniasis and Oroya fever.

Beetles

Some beetles act as intermediate host of the dwarf tapeworm *Hymenolepis diminuta*, an uncommon human parasite of minor importance.

Reduviid bugs

Reduviid bugs transmit *Trypanosoma cruzi*, causative agent of Chagas' disease, in South America. They are about 2.5 cm in length – much larger than bed bugs – and unlike them, they have wings. They are usually active at night, settling on the face of an unsuspecting sleeper to take a blood meal and to defecate. The infective trypanosomes are in the hindgut and the bitten person becomes infected by rubbing the bug's faeces into the irritating bite wound.

Fleas

Fleas are small blood-sucking parasites. They have laterally flattened bodies and lack wings. Well developed hind legs enable them to jump from host to host. Many fleas feed on man if given the opportunity. However, the species that is adapted for life on man is the human flea, *Pulex irritans*. It is common throughout the world. Female fleas of another species, *Tunga penetrans*, attack man once they have been fertilized. The fleas burrow into the skin, or under the toe-nails, of human host and are known as *jiggers*. The abdomen of the gravid female becomes grossly distended *with eggs*, causing pain, irritation and, sometimes secondary infection. Jigger fleas are common in dry, sandy soil, mainly in Africa and parts of Central and South America.

Human fleas are seldom implicated in the transmission of disease, but some other species are important disease vectors. Most notorious is the rat flea, *Xenopsylla cheopsis*, which is the most important, but not the sole vector of plague. Some forms of typhus are also transmitted by *X. cheopis* and other fleas.

Lice

Lice are wingless insects that undergo incomplete metamorphosis during their development. The ones that parasitize man are blood-sucking species with flattened bodies and short legs that are adapted to cling to hair. Body lice and head lice are considered to be variants of the same species, *Pediculus humanus*. Body louse, *P. humanus corporis* is somewhat larger than the head louse, *P. humanus capitis*, and there are other minor differences. A third species *Phthirus pubis* is quite distinct morphologically and is known as 'crab' louse.

Head lice are usually confined to the hair of the scalp, but body lice live in clothing covering the body, rather than on the skin. *Phthirus pubis* is usually found on pubic hair, but may also infest other hairy parts including eyelashes. All types attach their characteristic eggs (*nits*) to body hair and effective treatment involves removal of the *nits* as well as dealing with the adults.

Crab lice are not known to be involved in disease transmission, but body and head lice are classic vectors of epidemic typhus and relapsing fever. Treatment with insecticides such as permethrin, malathion and carbaryl may be effective, but resistance occurs.

Other Biting Flies

The tsetse flies, *Glossina* spp., are found in the so called 'fly belts' of sub-Saharan Africa, where they are responsible for transmission of trypanosomiasis in man as well as in cattle and other animals. Usually both male and female feed on blood. Other biting flies responsible for transmission of disease in Africa include species of *Chrysops* (deerflies or mango flies) which act as vectors of *Loa loa* and *Simulium* (blackflies) which act as vectors of onchocerciasis.

Midges

Biting midges are tiny flies. The females attack in swarms, usually in the evening, and may give rise to painful reactions. Like mosquitoes, they are mostly aquatic. One genus *Culicoides* spp. transmits filarial worms of *Mansonella* spp.

ARACHNIDS

This group includes spiders, scorpions, ticks and mites. They have four pairs of legs. They do not have wings, nor do they have antennae.

Ticks

Ticks are important vectors of human disease. They are of two types: ***hard (ixodid) ticks***, which have a chitinous shield on the back and ***soft (argasid) ticks***, which lack this feature. Ticks are obligate blood-feeders. They parasitize a wide variety of animals. Ixodid ticks transmit many rickettsiae of the spotted fever group as well as agents of Q fever, Lyme disease, tularaemia, babesiosis and some arboviruses.

Other Arthropods

Crustaceans (crabs and crayfish) are of interest in human medicine mainly as intermediate hosts of *Paragonimus westermani*, the lung fluke. Copepods (water fleas) are similarly important only as hosts of guinea worm, *Dracunculus medinensis* and the fish tapeworm, *Diphyllobothrium latum*.

Key Points

- Arthropods transmit infections by mechanical or biological transmission.
- Anopheline mosquitoes and culicine mosquitoes are of importance to human medicine.
- *Phlebotomus* species of sandfly is associated with widespread transmission in most of the parts of the world.
- Ticks are very important vectors of human disease.

ASSESS YOURSELF

MULTIPLE CHOICE QUESTIONS

1. Which type of vector is *Culex quinquefasciatus* for the transmission of *Wuchereria bancrofti*?
(a) Propagative (b) Cyclopropagative
(c) Cyclodevelopmental (d) Transovarian

2. Sandflies belong to which of the following classes?
(a) Insecta (b) Arachnida
(c) Pentastomida (d) Crustacea

3. Both males and females of which of the following genera of arthropods bite?
(a) *Phlebotomus* (b) *Glossina*
(c) *Crysops* (d) *Anopheles*

4. Which of the following arthropods bite during daytime?
(a) *Anopheles* (b) Reduviid bugs
(c) *Phlebotomus* (d) *Aedes*

5. Which of the following genera of arthropods can pass through ordinary mosquito net?
(a) *Phlebotomus* (b) *Culicoides*
(c) *Anopheles* (d) *Aedes*

6. In which of the following arthropods transovarial transmission of pathogens occurs?
(a) Sandflies (b) Ticks
(c) Head lice (d) Reduviid bugs

ANSWERS TO MCQS

1. c **2.** a **3.** b **4.** d **5.** a **6.** b