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## Part I

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Gargano and G.I. Zervakis

### General Aspects of Mushroom Fungi

Fungi are the most variable group of organisms, which demonstrate a great variety of morphology and life cycles. For being easily recognizable, fungi (as all living organisms) are given names and are classified into systems. Each species belongs to a genus, each genus to a family, etc. through order, class, phylum, and kingdom. In this way, the genus *Boletus* includes species like *Boletus edulis* and *Boletus satanas*; the genera *Boletus* and *Leccinum* are members of the family *Boletaceae*, while *Suillus* is a member of the *Suillaceae*; both families are members of the order *Boletales*, etc.

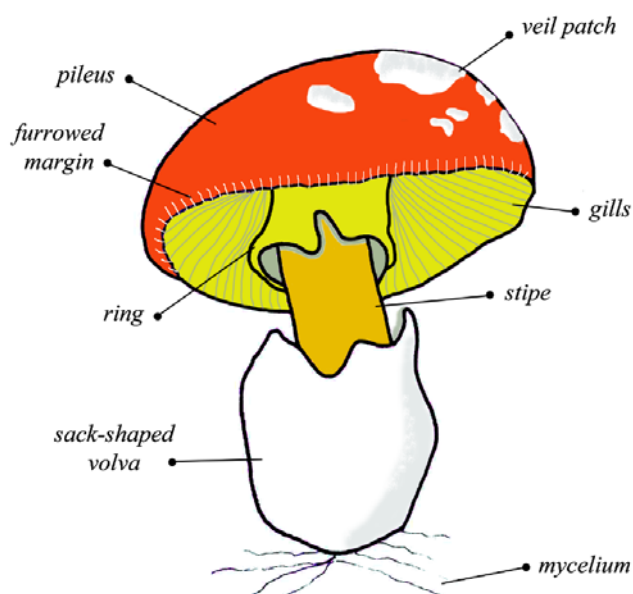
#### 1. *Morphology*

What collectors of wild mushrooms find in nature (on soil, stumps, rotting wood or other organic substrates) are merely **basidiomata** (sing. basidioma) of fungal species (previously called fruiting bodies or sporocarps). Their function is to form the spores and to aid their liberation. In fact, the main body of these fungi is situated below ground, in wood or in another, specific for the species substrate, and consists of a network of branched **hyphae** (thread-like structures), which form the fungal **mycelium**. The mushroom basidiomata and other structures (e.g. sclerotia) are composed of organized hyphae compacted into pseudotissues. At a certain stage of development and under favorable environmental conditions the mycelium may produce **basidiomata** (in basidiomycetes) and **ascomata** (in ascomycetes). In most cases they appear above the ground (or onto other substrates), but there are several species (both ascomycetes and basidiomycetes) which form ascomata and basidiomata under the ground.

The particular part on ascomata or basidiomata where the cells for sexual reproduction are formed (fertile part) is called **hymenium** and the whole surface that surrounds them **hymenial layer**. In the case of ascomycetes, the sexual spores are called **ascospores** and they are developed within specialized reproductive structures (sac-like cells) called **asci**, while in the case of basidiomycetes the sexual spores are called **basidiospores** and they are developed on top of specialized reproductive structures (club-shaped cells) called **basidia**, which bear sterigmata.

The morphology of the ascomata and basidiomata is characterized by a great diversity, and correct descriptions are important for the identification of both ascomycetes and basidiomycetes. The different groups of fungi appear on different substrates and have different strategies for spore dispersal: the ascomata and basidiomata may appear above or below the ground; those which are formed above the ground develop different types of associations with the substrate, from which they obtain nutrients. Others form symbiotic relationship mostly with woody and rarely with grassy plants. This kind of symbiotic relationship is known as **mycorrhiza**.

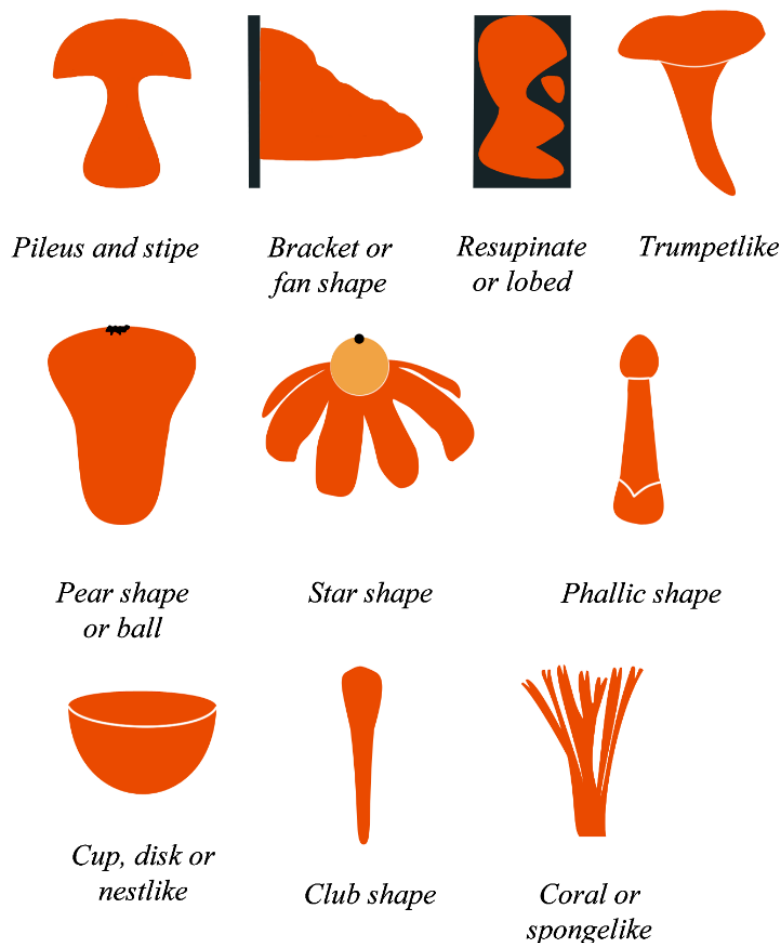
The diversity of ascomata and basidiomata is enormous, e.g. there are species whose basidiomata are relatively simple and grow on the substrate (wood or soil).



**Fig. 1.** Basidioma of a gilled mushroom (*Amanita caesarea*); note the veil patches (remnants of the universal veil), the stipe ring (a remnant of the partial veil), and the sac-shaped volva (a remnant of the universal veil) (Drawing by T.T. Denchev).

In most basidiomata, there are well-shaped **pileus** (also called **cap**) and **stipe** (also called **stem**), and the hymenial layer is raised above the substrate (Fig. 1). In other cases the differentiation aims towards better protection and the hymenium remains enclosed in the basidioma until it is fully mature (this is the case with the so called gasteromycetes or gasteroid fungi, e.g., *Calvatia gigantea*).

The following main **types of shape** can be recognized among the basidiomycetes and discomycetes (i.e., a former taxonomic class of ascomycete fungi which includes all of the cup, sponge, brain, and some club-like fungi) (Fig. 2):



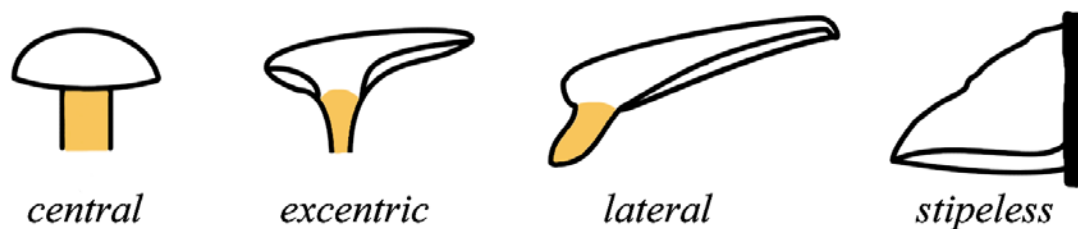
**Fig. 2.** Different shapes of basidiomata and discomycete apothecia (T.T. Denchev).

- **pileus and stipe** (e.g., *Agaricus*, *Boletus*);
- **bracket or fan shape** (e.g., *Fomes*, *Trametes*);

- **resupinate** or **lobed** (e.g., *Serpula*, *Tremella*);
- **trumpet-like** (e.g., *Craterellus cornucopioides*);
- **pear shape** or **ball** (e.g., *Lycoperdon*, *Calvatia*, *Scleroderma*);
- **star shape** (e.g., *Geastrum*);
- **phallic shape** (e.g., *Phallus*, *Mutinus*);
- **cup, disc** or **nestlike** (e.g., discomycete apothecia, *Cyathus*, *Crucibulum*);
- **club shape** (e.g., *Clavariadelphus*, *Macrotyphula*, *Typhula*);
- **coral** or **sponge-like** (e.g., *Ramaria*, *Hericium*, *Sparassis*).

For the fungi that produce basidiomata with pileus and stipe, the variability of the shapes, colours, sizes, structures, and the macrochemical reactions of the pileus, stipe and flesh/context to specific reagents, as well as the presence or absence of a veil, are important taxonomic characters.

The **pileus** ensures the connection of the hymenial layer and its protection. Its diameter may largely vary in different species, 0.5–20 (–60) cm in diameter.



**Fig. 3.** Types of pileus to stipe attachment.

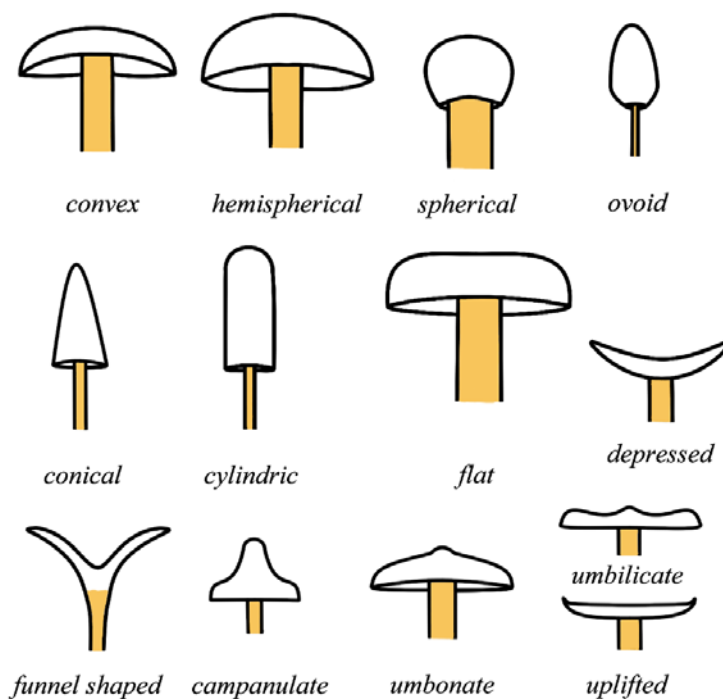
Depending on its position, the pileus may be connected to a stipe (with **central**, **excentric** or **lateral** attachment) or directly to the substrate (**stipeless** attachment) (Fig. 3).

The upper surface of the pileus may be **shiny** or **mat**; **dry**, **wet** or **sticky**; **glabrous**, **scaly** (appressed or uplifted), **velvety** or **finely powdered**. Sometimes the entire surface may be **veined**, **wrinkled**, **rugged** or **uneven**.



**Figs. 4 & 5.** *Suillus luteus* and *Clitocybe gibba* (Bulgaria, the Rhodopes; C.M. Denchev).

The pileus cuticle may **peel off easily** (the case of *Suillus*, Fig. 4) or **with difficulty** or may not be peeled off at all.



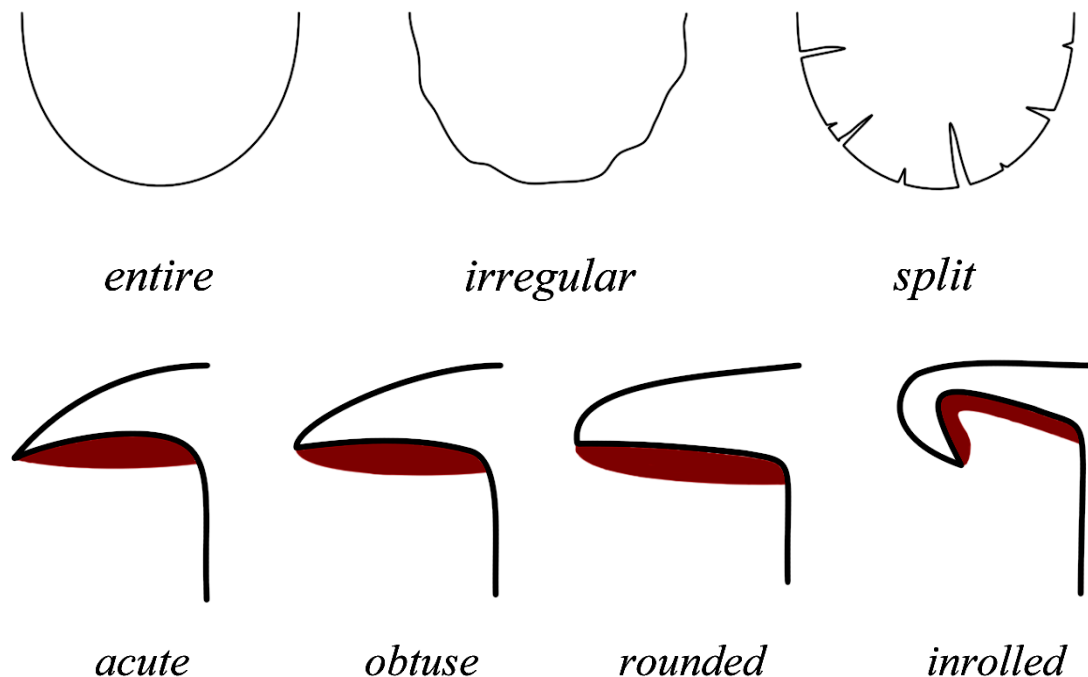
**Fig. 6.** Different types of pilei shapes.

According to its shape the pileus may be **convex** (domed upward, cushion-shaped), **hemispherical**, **spherical**, **ovoid**, **conical**, **cylindric**, **flat**, **campanulate** (bell-shaped), **umbonate** (with a central bump), **papillate** (if the central bump is very small and

pimple-like), **depressed** (with a depression – Fig. 5), **umbilicate** (with a depression with a small bump or pimple in the centre), **funnel shaped** (with a very deep depression), **uplifted**, etc. (Fig. 6).

The pileal margin may be **entire**, **irregular** or **split** (Fig. 7); with or without **radial streaks**; **thin** or **thick**; **curved outwards**, straight (**acute**, **obtuse**, **rounded**) or **inrolled** (Fig. 7); in the beginning it is often inrolled and later becomes flat. The hymenophore usually **reaches the pileal margin**, but it is also possible that it does **not reach it** (such margin is called "sterile").

The hymenophore may be composed of **gills** (e.g., in *Agaricus*; *Amanita*), **tubes** (e.g., in *Boletus*, *Suillus*), **spines** (e.g., in *Sarcodon*, *Hydnum*), **ridges** or may have other forms.



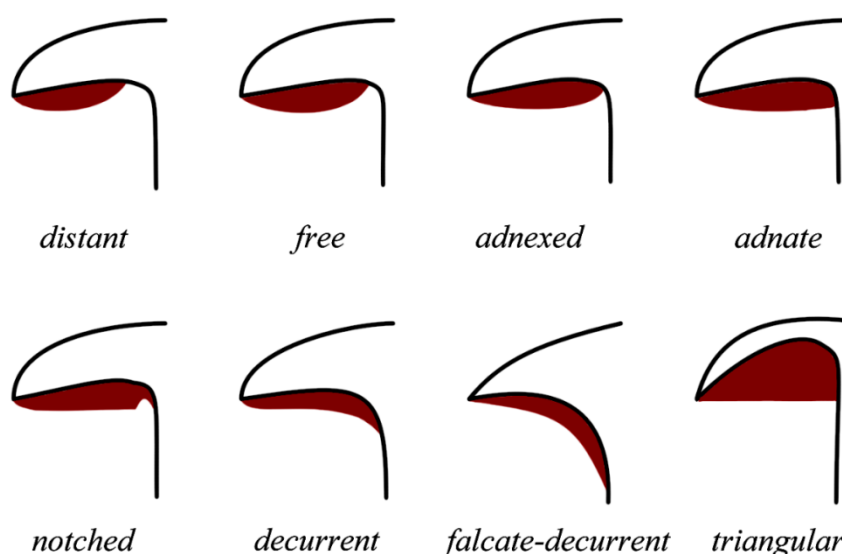
**Fig 7.** Various types of pileus margins.

The gills attachment to the stipe (Fig. 8) may be (i) **free** (not reaching the stipe; a circular gap is visible around the top of the stipe where the pileal underpart can be seen), **distant** (when the distance between the gills and stipe is large), or **remote** (when the distance is even larger); (ii) fused – **adnexed** (narrowly attached to the stipe), **adnate** (broadly attached), **notched** (appearing as if a notch was removed from

the lower part of the gill where it attaches to the stipe), or **triangular**; (iii) **decurrent** (running down the stipe top) or **falcate-decurrent** (decurrent gills which arch upwards and outwards towards the pileal margin).

In species with gilled hymenophore, the gill edges may be **entire** (smooth and even), **dentate** (having toothlike pointed projections), **serrate** (having sharp teeth like a saw), **crenate** (coarsely jagged) or **ciliate** (fringed) (Fig. 9).

The basidiomata of some species may have lactiferous hyphae (e.g., the pileus of the genus *Lactarius* has hyphae which contain white or coloured milky latex). The colour of the flesh varies, but most often it is white. The colour of the flesh in many species may change when exposed to air (the so called **auto-oxidation**); this is an important character for identification. The flesh texture may be soft, leathery, gelatinous, corky, woody, etc.

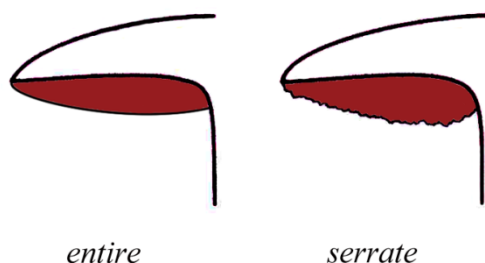


**Fig. 8.** Types of gill attachment to stipe.

The **stipe** is in most of the cases the non-reproductive part of the basidioma and it supports the pileus, being differently rigid in various species (Fig. 10). The pileus is connected **centrally** or **eccentrically** to the stipe. In species which the pileus is directly attached to the substrate, the stipe is lacking. The stipe is composed by vertically arranged and tightly packed and interconnected hyphae. It raises the

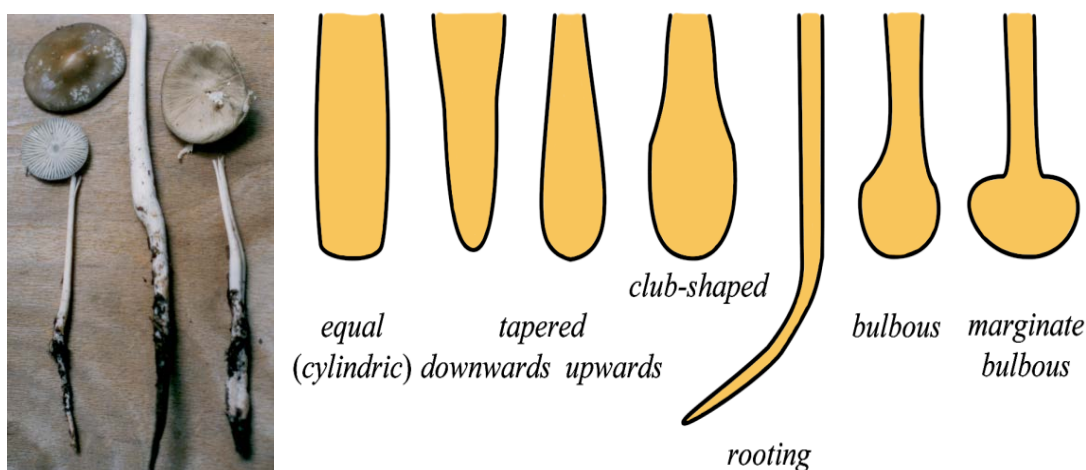


hymenophore above the soil (or other substrate, specific for a particular fungus), thus facilitating the better liberation and dispersal of the spores.



**Fig. 9.** Shapes of gill edges.

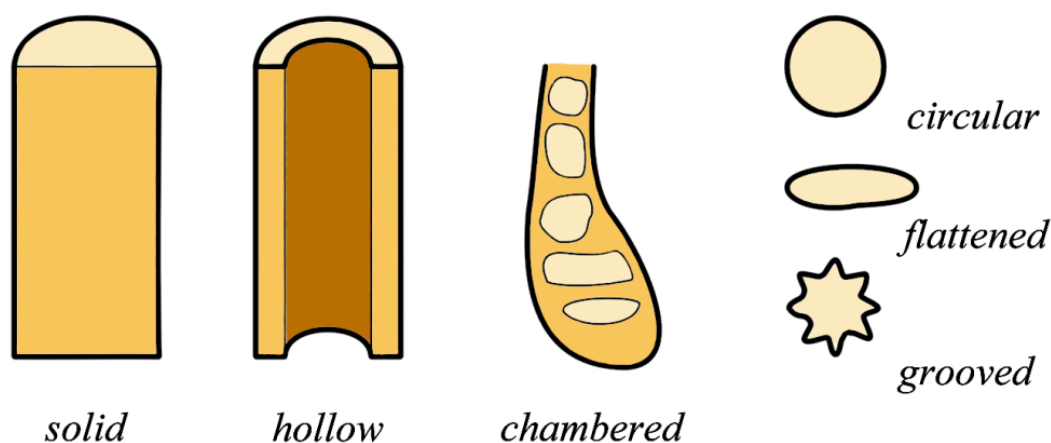
By its shape the stipe may be **spherical**, **egg-shaped**, **paddle-shaped**, **inversed paddle-shaped**, **cylindrical**, **spindle-shaped**, **tapering towards the base**, **rooting** (Fig. 10). In species with heavier pilei, the stability is ensured by the top to base gradual widening of the stipe or by its bulbous swelling at the base part.



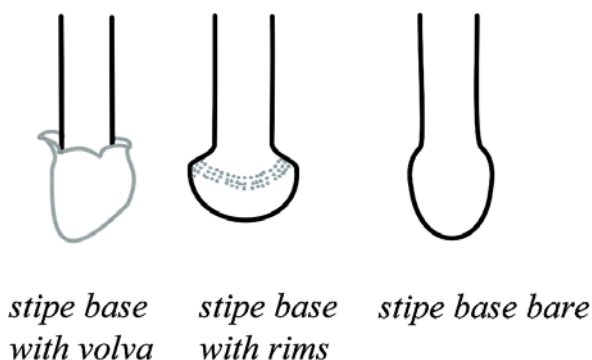
**Fig. 10.** Rooting stipe of *Xerula radicata* (Bulgaria, the Rhodopes; C.M. Denchev), and various types of stipe-root shapes.

The stipe may be **straight** or **curved**, and **solid**, **hollow** or **cavernose** (chambered – with separate cavities), and in cross section – **circular**, **flattened** or **grooved** (Fig. 11). By its texture, surface or viscosity, the stipe could be: **fragile** or **leathery**; **soft**, **tough** or **woody**; with **smooth**, **scaly**, **warty**, **fibrillose** or **furrowed** surface; **dry**, **sticky** or

**viscid.** Depending on the presence of remnants of universal or partial veil, the stipe may or may not bear the so called **ring** – remnant of partial veil (Fig. 12); and may be with or without **volva** (or bag – Fig. 13), **collar** or **rims of warts** (Fig. 13).



**Fig. 11.** Sections through different types of stipes.



**Figs. 12 & 13.** *Amanita muscaria* (Bulgaria, Mt. Vitosha; C.M. Denchev), and stipe base with volva, with remnants of universal veil or bare.

For the pileus, as well as for the stipe, it is important to evaluate the colours properly, for which the use of standard colour charts is recommended.

In basidiomycetes the following **major basidiomata types** are distinguished:

- **Open** (without a veil) or **semi-open** (with a veil) basidiomata, consisting of pileus and stipe or of a pileus only. The **veil** may be universal or partial. The **universal veil** (velum universale) is a membranous or leathery bag, which completely encloses the

young basidioma. During growth, the universal veil cracks and tears, leaving remnants in the stipe base. These remnants may be in the form of a bag-like volva, collar or rims of warts or scales. Other remains of the veil may appear on the pileus in the form of powdery grains, scales or warts of different shape, size and placement. The **partial veil** (velum partiale) is membranous, thread-like or cobweb, which in young stage connects the pileus margin with the upper part of the stipe, covering the spore-bearing surface, situated on the lower surface of the pileus. With the mushroom growth, the partial veil tears and part of it remains as a lasting or disappearing ring, usually placed on the upper part of the stipe. Sometimes on the pileus the presence of scaly or thread-like remnants of the partial veil could be observed.

Depending on the **presence or absence of veil**, fungi of this group may be:

- species with universal and partial veil;
- species with a universal veil only;
- species with a partial veil only;
- species without universal and partial veils.
- **Closed** basidiomata of spherical, egg-shaped, tuber-like or pear-like shapes, enclosed by a protective layer (peridium). Here belong the so called gasteromycetes.
- Basidiomata with **shrub-like** (coral-like) and **bat-like** shapes. The basidiomata may be branched with shrub-like shape (*Ramaria*) or unbranched, bat-like (*Clavariadelphus*). The spore-bearing layer is situated on the external surface in the upper part of the basidiomata. Some of the species in this group are edible, but this group is not included in the present textbook.

Of special interest for the identification of fungi is the **colour of the spores** as determined after obtaining their print on a piece of paper. It may belong to one of the following groups:

- white to cream,
- pinkish to red,
- ochraceous to clay,
- reddish brown,
- purplish,
- black.

Of course, there might be some exceptions from the above groups (e.g., ascomycetes with green spores), but they are not included in this textbook. In order to obtain a good spore print, one needs a fresh mushroom specimen (it cannot be produced from a dried specimen) and then to act as follows: Place the cap on sheet of paper and cover with glass. Leave for several hours or overnight to allow the spores to be released. On inspection, the print left by the spores on the paper will mirror the spaces that exist between the gills. Some manuals even recommend that the cap is placed half on white, half on black paper, especially when the approximate colour of the spore print is not known in advance. Thus, light-coloured spore prints will show up better on darker paper and vice versa. In addition, spores release may be facilitated if a drop of water is placed on the cap before covering it.

In the ascomycetes, included in the textbook, the **ascomata** are either born above the ground apothecia, or underground, closed, tuber-like bodies.

The ascoma of the morels (*Morchella* spp.) is an **apothecium**. It comprises of spherical, spherical-elongated or conical pileus with a honeycomb-like outer surface, and a rigid, stipe-like structure. On the upper surface the hymenial layer is situated which contains the structures where the spores are born (called **asci**).

The ascomata of the truffles (*Tuber* spp.) are hypogeous (formed into the soil), and usually subglobose.

## 2. *Collecting fungi for identification*

Collecting fungi for identification is an important stage of this process and attention must be paid. For collecting specimens one will need some consumables and suitable equipment. First of all suitable paper bags or plastic boxes are needed to store the specimens when collecting in the field. Plastic bags are not recommended as mushrooms tend to disintegrate or change features. Alternatively aluminum foil may be used to carefully wrap the basidiomata, gathered for identification. If the specimens could not be processed on the same day, they might be kept for a certain time in a fridge, but freezing is not suitable. When collecting mushrooms for identification,

basidiomata and ascomata of different ages should be collected, when this is possible, as in many fungi there might be striking differences between young and mature mushrooms. As in some of them certain characters might be ephemeral and disappear in a few hours, taking photographs in the field is most recommended.

Attention to the habitat and especially on the surrounding vegetation should be paid, since the knowledge about this may greatly aid the identification.

When the specimens are taken home, the next important step is to carefully take notes on vital characters, which are important for achieving correct identification. Taking notes of as much characters as possible, e.g., features of the pileus (size, colour, surface, shape, margin), stipe (size, shape, colours), veil (presence or absence, type, remnants), flesh (colour, colour changes, surface, smell, in certain cases the taste might be important, but one should never taste any unknown mushroom as to avoid poisoning), hymenophore (type, colour, colour changes, shape, attachment, etc.), is very important for subsequent identification as explained in *Morphology*. In many cases, for arriving at the correct identification, the micromorphological characteristics of specimens should be examined with the aid of a microscope. Anatomical features (e.g. spores, cystidia, basidia, basidioles, hyphal systems, hyphal walls, septations, hyphal branching, hyphal inflations and specialized hypha) could be thus evaluated. In addition, the use of chemical reagents is often necessary, i.e. 95% ethanol (v/v in water), 3% potassium hydroxide, 5–10% ammonium hydroxide, Teepol, acetocarmine, chloral hydrate, Congo red, cotton blue, cresyl blue, fuchsin, guaiac, hydrochloric acid, Melzer's reagent, methylene blue, sodium hydroxide, sulphobenzaldehyde and sulphuric acid. Finally, a spore-print should be prepared. After all this is done, one may proceed with the identification using suitable guides and identification handbooks. In many cases precise identification may be only achieved after the study of fungal specimens under a microscope.

In certain cases, specimens may be stored, especially when a scientific study is intended. Specimens are dried either in open, or in dryers, and it is recommended that drying temperature do not exceed 45 °C. After drying the samples are kept in a dry place, in paper packages, labelled with the name of the species, if known, the locality of collection, the date and the name of the collector.