KOR. JOUR. MICROBIOL. Vol. 8, 41~51 (1970)

酵母의 生態學

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Ecology of Yeasts

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Introduction

Yeasts are widely distributed in nature. However our knowledge of the occurrence and distribution of yeasts tells us that their natural habitats are rather specific. As many of the living forms are restricted their living places by the ecological relationships with the environment, so are the yeasts. Undoubtedly studying of yeasts in such ecological aspects will reveal their roles played in nature and both of their beneficial effects and harmful affliction to our living.

As Lund (33) has described, in the temperate zones where four different seasonal changes are apparent, yeasts are cycling from substrates to substrates in nature as the season changes.

In late summer abundant yeasts cells are supplied to the soil from various substrates, such as wind fallen fruits and decomposing fleshy fungi where yeasts prosper during summer. The yeasts on the surface of the ground are carried into some depth of the ground by seeping water, animals and tilling of soil etc.

Yeasts are capable of reproducing in the soil at any rate under certain conditions. However the greatest significance of soil lies in its provision of a reservoir in which yeasts can survive adverse periods like winter. Laboratory tests (33) have shown that yeasts withstand low temperature and can remain viable for at least 6 months.

Also some yeasts which are consumed by herbivorous animals with their fodder pass unhurt through the alimentary canal and fall in the dung of horses and cattles. Yeasts also appear to be capaple of hibernating in dung.

Beehives are also one of their hibernating place in winter. Honey bees were examined early spring before they had started flying about and yeasts were found.

Thus yeasts hibernate in adverse winter in soils, dung and beehives. In spring then yeast cells may be conveyed into the air from the surface of soil and dung. From deeper layers of the soilboring animals may be able to carry yeast cells up to the surface, also by tilling of the soil. Larger quantities of yeast cells were found on the surface of the ground in spring than in summer. Probably in summer strong ultraviolet and dessication etc. would result to the decreased number of yeasts. By the wind, animals and heavy showers yeast cells then may be conveyed to substrates. Bees also may carry yeast cells to flowers to flowers, while bees themselves were good habitats for yeasts. Currents of water carry yeasts into lakes and ocean.

These yeasts then multiply vigorously in exudates of trees, while flowers usually seems to contain only small amount of yeast cells.

From summer to autumn, substrates which offer conditions of growth to yeasts are exudates of trees, bees, flowers, fruits and fleshy fungi. While yeasts are prospering, they are more vigorously disseminated among those substrates. Besides by the movement of air and water, *Drosophilla* flys which feed on yeasts from their own nutritional requirements carry yeasts among substrates like spoilaging fruits, also among tree exudates for oodepositing because the larvae also feed on yeasts. Bark beetles also disseminates yeasts into their tunnels under bark of trees.

Thus prospered yeasts all go into soil in autumn and hibernate in winter to repeat their cycle again the next year. While they are thus circulating in nature, of course, their occurrence in various ecological niches could be detected by various methods of ecological studies. Some of those results, which actually revealed the above aspects of yeast ecology would be summarized here.

In ecological studies, the flora always consists of two groups of yeasts, that is, the one which has ecological significance with the substrate proper and the other which is only an occassional occurence. It would be tried in this discussion that they are treated separately. In the discussion of various habitats of yeasts in nature only those yeasts which are ecological significances are discussed and all the occasional yeasts present in various substrates are summarized as a separate group.

Media for enumeration of yeasts

Previous works on ecology of yeasts until Hertz & Levine (28) started to work out a selective medium for yeasts were in general exclusively based on results obtained by means of enrichment cultures. Only in exceptional case countings of yeasts were made, presumably because of the difficulty in avoiding overgrowing of accompanying organisms on the usula nutrient media. When the occurence of yeasts in enrichment cultures of a substratum is asurtained, this need not, however, mean that the substratum in question is a habitat for them, as it may be a case of quite an accidental occurence of yeast cells which have been carried to the substratum by the wind or insects or in other ways.

Therefore significant ecological studies may be considered to be started after the invention of selective media of yeasts for direct plate counting method.

Hertz & Levine (28) found that diphenyl (0.01%) when added to agar media suppressed molds without affecting yeast growth. Na-propionate seemed less suitable for separation of yeasts and molds than diphenyl. 0.3% of sodium propionate inhibits molds but also yeasts. With 0.2% yeasts are less damaged but molds were not enough inhibited.

Mrak & Pheff (36) found that 0.25% sodium propionate quite useful for separation of yeasts from molds.

Miller *et al.* suggested that oxgall (2%) added to potato dextrose agar facilitated the isolation of yeasts from soil by inhibiting bacteria and molds.

Wickerham (68) used technique for inhibition of mold growth by preventing the entrance of air into enrichment culture. He covered the plating media with sterile mineral oil. This method favored the development of fermentative species.

Hesseltine *et al.* (29) found that addition of aureomycin to enrichment medium facilitated the isolation of yeasts from soil and other materials high in bacteria.

Miller & Webb (41) succeeded to isolate yeasts from soil with the aid of acid, rose bengal (0.003%) and oxgall (1%).

Etchells *et al.* (17) found that acidified dextrose agar with 0.35% sodium propionate is the most suitable for examination of soil yeasts. 0.02% diphenyl failed for inhibition of molds.

Lund (33) in his studies of yeast ecology used 0.25% sodium propionate added wort agar. He found this was sufficiently effective in suppressing mold to permit counting and isolation of yeasts with fair certainty.

Beech and Carr (2) in studying yeasts in apple juices and ciders found 0.01% diphenyl in malt agar good for suppressing mold while permitting satisfactory growth of yeasts.

It seems that some variations of effectiveness of those selective media exists depending upon conditions of test and kinds of yeasts.

Habitats of yeasts in nature

A. Flowers:

Lund (33) extensively studied yeasts occurring in various flowers near Copenhagen. Predominant yeasts were;

Cryptococcus albidus
Torulopsis famata
Candida reukaufii

Etchells *et al.* (18) found prodominant yeasts in cucumber flowers as;

Rhodotorula glutinis
Rh. flava

Capriotti (33) examined flowers in Italy, most common species were;

Candida reukaufi
C. guilliermondii

Generally it seems that no relationship between type of yeasts and type of flower. Though all the yeasts isolated from flowers are only imperfecti.

Lund(33) confirmed that yeasts occurring in flowers are mostly inoculated by insects visiting flowers.

B. Fruits: The predominant yeasts so far known in fruits are;

1. Souring figs, Mrak *et al.* (37)
   Saccharomyces cerevisiae
   Candida krusei
   Pichia kluyveri
   Kloeckera lindneri
   Hanseniaspora melligeri
   Torulopsis stellata
2. Plums, De Migoya (12)
   Hansenula anomala
   Schizosaccharomyces niger
   Debaryomyces sp.
   Zygosaccharomyces sp.
   Hanseniaspora apiculata
3. Apples, Clark *et al.* (1954) (5)
   Candida malicola
   Cryptoccus albidus
   Cr. laurentii
   Cr. neoformans
   Torulopsis famata
4. Berries Shihta *et al.* (54) found prospering in juniper berries and manzanita berries.
   Kloeckera apiculata
   Rhodoturula glutinis
5. Lund (33) extensively studied fruits of 13 plant species. According to his results the most predominant species are;
   Kloeckera apiculata
   Candida pulcherrima
   Torulopsis famata
   Candida reukaufii
   Candida parapsilosis
6. Dried fruits, Mrak *et al.* (38) examined

yeasts occurring in dates. Predominant yeasts were;

Zygosaccharomyces japonicus var. soyer
Hanseniaspora melligeri
Candida chalmersi
Zygosaccharomyces barkeri

Van der Walt (57) found in Agrican dried figs.

Zygosacch. cavarae var. *beanverie*
Zygosacch. mandschuricus

are the chief parts of yeasts.

It is generally hard to draw any conclusion to all of the fruits together. But it can very vaguely be said that in fresh fruits *Kloeckera*, *Hanseniaspora* are almost common. In dried fruits *Zygosaccharcomyes* are characteristics.

C. Honey and Syrup

Fabian and Quinnet (25), Lochead and Heron (34) found the osmophilic yeast flora in honeys as;

Zygosaccharomyces nusbaumeri
Zygosacch. japonicus
Zygosacch. mellis
Zygosacch. barkeri

English (22) found the chief osmophilic yeasts in malt extract as;

Saccharomyces rouxii

Ingram (32) also found *Zygosaccharomyces* sp. in concentrated orange juice.

D. Trees and Tree exudates

Trees are one of the most important natural habitats of yeasts. Wickerharm (68) discussed the evolution of the genus *Hansenula* and suggested that the association of yeasts with trees protects those physiologically weaker yeasts by eliminating more vigorous and therefore more competitive microorganisms from environments.

Phaff *et al.* (44. 6) extensively studied yeast flora of the natural breeding sites of some species of *Drosophilla* of California. The most predominant yeast species on the slime flux of *Abies concolor* were found as;

Hansenula mrakii
Debaryomyces fluxorum
Pichia silvestris
Pichia pastori

The interesting facts are that the *Drosophilla* flys which feed on yeasts spare those yeasts in slime fluxes just for their larvae feeds and not for their own.

Lund (33) also studied yeast flora near Copenhagen found quite different picture of ecology. Depending upon trees, climate probably the yeast flora on tree exudates may quite be different. According to Lund (33) the chief species on various tree exudates were as;

Toulopsis molischiana
Candida pulcherrima
Candida krusei
Candida mycoderma

Van der Walt (58) found that

Endomycopsis wickerhamii

in the larvae gut frass of *Cycadales* trees in Africa.

E. Fleshy Fungi

Decomposing fleshy fungi are good habitats of yeasts. Anderson (1) found species of *Saccharomyces*, *Crytococcus*, and *Rhodotorula* though the predominants were not clear.

Carson *et al.* (6) isolated *Candida humicola* from *Clavaria* and *Pleurotus* fungi.

Lund (33) also examined fleshy fungi near Copenhagen and found predominant yeasts as;

Candida parapsilosis
C. mesenterica
Torulopsis colliculosa
Kloeckera apiculata

F. Animal

Rettger *et al.* (52) found that the ingested bakers yeasts showed rapid and extensive destruction in the alimentary tract of human though there were always comparatively small numbers of viable cells.

Lund (33) examples of horses and cow dung which had just been dropped in a field. There was a comparatively large number of yeast cells which must originate from the fodder. Predominant yeasts were;

Candida krusei

C. parapsilosis

C. mycoderma

It seems that most of the wild yeasts pass the alimentary canal in an uninjured state contrary to bakers yeasts.

Parle (51) studied extensively yeasts isolated from the mammalian alimentary tract. He considered only the following four species to the true intestinal flora.

Sacchromycopsis guttlata

Torulopsis pintolopesii

Candida albicans

S. guttlata were always found in rabbits, *T. pintolopesii* in all guinea pigs, 73% of mice and 66% of rats examined. *C. albicans* were dominant in sheep, hedgehogs and opossums and the widely distributed of the species isolated.

According to Phaff *et al.* (45) *Saccharomycopsis guttlata* is intimately associated with rabbits. Young rabbits are needed to be inoculated by this yeast for their normal growth.

According to Van Uden *et al.* (57, 58) main intestinal saprophyte yeast of swine was;

Candida sloffii

and the facutative intestinal saprophytes are considered as;

Candida krusei

Saccharomyces cerevisiea

Pichia membranefaciens

The intestinal yeast flora of free living hippopotami wart hogs and bushpigs were more or less similar to the swine. Species of *Candida* were the most common with the occasional isolations of *Debaryomyces*, *Trichosporum*.

Reiersöl (53) reported the most frequent occurence of *Candida albicans* and *Candida mycoderma* in the intestinal tract of human patients.

G. Insects

Insects are not only important in nature in having a role to disseminate yeasts among various substrates but also some insects are closely associated with peculier yeasts at specific ecological niches of yeasts. Notably well studied examples of yeasts and insects relationships are *Drosophila* flys and bark beetles.

It has been noted that yeasts are necessary for normal growth of *Drosophila*. According to Shiheta *et al.* (54) yeasts are important in understanding some of the forces of natural selection to which the natural population of *Drosophila* are expose.

Since from time to time yeasts in a given area may vary, the growth and development of one type of fly may be favored at one time and some other type at another time and also variation in the intestinal flora of yeasts occurs.

According to Phaff *et al.* (46, 47) at low altitude of Yosemite region of California, the most common intestinal yeast flora of *Drosophila obscura* and *Drosophila pinicola* were;

Saccharomyces montanus

S. veronae

S. cerevisiea var *tetrasporus*

S. drosophilarum

Hansenula angusta

Kloeckera apciulata

while at high altitude *Drosophila pinicola* only had mainly bacteria.

Dobzhansky *et al.* (13) found different attraction of species of *Drosophilla* to different species of yeasts. Phaff *et al.* (46, 47) found that in the hills *Drosophilla obscura* can feed on *Saccharomyces veronae* while *Drosophilla melanogaster* has no ability to find this wild yeast.

The intestinal yeast flora may reflect that the yeast flora of the area. Hedrick *et al.* (30) found the intestinal yeast flora of Hawaii as;

Hansenula anomala
Candida krusei
C. monosa
C. intermedia

This is somewhat different from that of Phaff *et al.* (46, 47) found in California.

Bark beetles, their host trees and yeasts are closely interrelated. Miller & Mrak (42) found the chief yeasts associated with dried-fruit beetles in figs as;

Candida krusei
Hanseniaspora valbyensis
Torulopsis carpophila

Shiffrine & Phaff (55) found the predominant yeasts associated with bark beetles, *Dentroctonus*, and *Ips* in pine trees as;

Pichia pini
Hansenula capsulata
Candida silvicola
Candida curvata

However the pine tree, *Pinus seffreyii* did not have *Pichia pinii.*

Phaff & Yoneyama (48) reported that the bark beetle *Scolytus* in fir trees associated with

Endomycopsis scolyti

and yeasts associated with bark beetle of western hemlock tree as (49);

Hansenula capsulata
Sporobolomyces singularis
Candida oregonensis
Torulopsis candida

Owem & Mobley (43) reported that

Torula acidophila

is associated with the American cockroach *Periplaneta americana.*

Bees are insects which diseminates yeasts in nature. But they themselves are a good habitat of yeasts. Lund (33) examined 29 samples of bees and wasps found the predominant yeasts as;

Candida reukaufii
C. pulcherrima
C. krusei
C. guilliermondii
Kloeckera apiculata
Torulopsis bacillaris
Hansenula sp.

Hajsig (31) found

Torulopsis apicola

as the normal flora of bees in Yugoslavia.

Van der Walt (50) found in the larvae feed of the South African bumble bee yeasts of species, *Torulopsis*, *Candida*, *Saccharomyces*, *Pichia*.

H. Grains

Del Prade *et al.* (7) found the sole yeast flora of stored nice seed was;

Candida pseudotropicalis

Tennisson (56) reported yeast flora of stored rice as;

Endomycopsis chodati
Hansenula anomala
Pichia farinosa
Candida krusei

Lund (33) found in barleys yeasts of species of *Rhodotorula*, *Sporobolomyces* as dominant and much less of *Hansenula*, *Torulopsis*, *Candida*.

I. Wood

Rennerfelt and Goidanich *et al.* (36) studied yeasts in wood pulp in Swedish paper mills. Species of *Rhodotorula*, *Torulopsis. Candida* were found.

Carlson *et al.* (6) and Shihata & Mrak (54) found in various decaying woods species of yeasts, *Hansenula*, *Pichia*, *Candida*, *Sporobolomyces*.

J. Milk

Emmons (18) found *Cryptococcus neoformans* responsible for highly viscid mucoid milk from a severe outbreak of Bovine Mastitis.

However most of the yeasts found in milk are contaminants of soil and air and all are also lactose fermenting. Di Menna (15) probably studied most extensively yeast flora of milk. He found the most predominant yeasts in pooled raw milk as;

Saccharomyces cerevisiae
Pichia fermentans
Candida mycoderma
Candida krusei
C. macedonensis
C. parapsilosis

K. Brines

Brines of various natural fermentations are a good substrate especially of those osmophilic yeasts.

Mrak and Bonar (39), Etchells & Bell (19), Etcheel *et al.* (20, 21) Costilow (7) and Costilow *et al.* (8) studied yeast flora in brines of cucumber and meat fermentation. Summarizing those results in those brines generally the following yeasts are predomenant.

Debarypmyces membranaefaciens
D. klockeri
D. membranaefaciens var *hollandicus*
Torulopsis holmii
T. carloliniana
T. rosei
Brettanomyces versatilis
Brett. sphaericus
Hansenula subpiliculosa
Zygosaccharomyces halomembranis
Zygo. globiformis

L. Wine and Cider

According to Castelli (9) in winery fermentations in France the always occurring yeasts in addition to other occasssional yeasts were;

Saccharomyces ellipseudes
Kloeckera apiculata

Van der Walt (60, 61) found that yeasts causing turbidity in South African table wines as;

Brettanomyces intermedius
Brett. schanderli
Saccharomyces acidifaciens
S. oviformis
S. cerevisieae
Pichia membranefaciens

Clark *et al.*(5) studied cider yeasts and found

Pichia membranefaciens
Saccharomyces oviformis

and others which are all not found on the apples.

Challinor (10) found in the fermentation of apple juice frequent apiculate yeasts and occassionally

Saccharomycodes ludwigii

type yeasts.

M. Soil

Bouthilet (3), Lund (33), Di Menna (16) and Capriotti (11) studied extensively yeast flora of soil. However surveying of their literature shows the difficulty to draw any general ecological conclusions. As has been pointed out in the introduction, the amount of yeast cells in the soil is undoubtedly first of all conditioned by the supply of substrate like fruits and fleshy fungi, exudates of trees, and insects, which may contain an abudance of yeasts and which add nutrient substances suitable for the development of yeasts to the soil. Therefore it is hardly to say that certain types of soil should be more favorable to yeasts than others.

Of course there are some yeasts which have been isolated only exclusivly from soil, the genera, *Schwanniomyces*(26, 62), *Kluyveromyces* (63) and *Lipomyces* (33). However from the reason stated above it is not clear whether their original habitats are soil or they have other own habitats which is not known.

N. Ocean

According to Zobell (69) yeasts were generally found in all samples regardless of the distance from the land. Whether the yeasts are originated from the land is not clear. However according to Van Uden and Zobell (66) yeasts were rather found in sea water samples collected over coral and algal growth.

Candida reukauffi
Pichia farinosa
Saccharomyces estuarii

were the isolated yeasts. Fell (27) also isolated *S. estuarii* from sea water sediment.

Phaff *et al.* (50) examined yeasts in shrimp, *Peneaus setiferus*. Species of *Torulopsis, Rhodotorula, Triclosporon, Candida, Hansenula* were identified. But no *Metschnikowiella* species were found.

The genus *Metschnikowiella* is well known than it has the habitats especially in brine shrimps though according to Van Uden *et al.* (67)

Metschnikowiella zobellii
M. krissii

were also found in sea water, fish guts and algae.

O. The others

Some yeasts are very rare in their occurrence.

Vander Walt (64) isolated

Endomyces reesii

in cold-water retting of *Hibiscus cannabinus.*

Nadsonia is exclusively isolated from tree exudate of cool climate zone (35). *Saccharomycodes* is also rarely found in trees (35).

Pityrosporum ovle is in planting seales from *Seborrhea oleosa* (dandruff) (24).

Sporobolomyces is usually found in plant leaves (4).

Most of the yeasts of the subfamily *Nematosporoideae* are plant pathogenics (4).

Nematospora coryli is the cause of disease in citrus, tomatoes, cotton balls, coffee beans, and beans.

Ashbya gossypii are found in fruits and seeds of similar host plants as the *Nematospora.*

Spermophthora, Eromothecium are in young lint fibers of cotton. It is reported that *Ashbya* is transferred by the insects, *Dysdercus, Leptoglossus, Nezara, Autestia.*

The genus *Wickerhamia* is found in squarrel droppings (35).

Yeasts of the widely distributed

While surveying the occurrences of yeasts in natural habitats it seems evident that some yeasts are especially noted in their wide distribution in various substrates. Though in each specific substrate they don't contribute as the predominant member of flora, they may be peculier in that they are rather widely distributed and rather not specific to any substrate alone. If the free living yeasts are too more phylogenetically developed yeasts as Wickerham (68) discussed, then this group of yeasts are the most likely one.

Also if a yeast species is isolated frequently both from flowers and insects, it may be reasonable to draw conclusions that the yeast in flowers are inoculated by insects. Also it may be noted that most of the yeasts in natural substrate also occur in soil.

Based upon such an idea the most widely distributed yeast species are examined and their occurrences among various substrates are summarized as the following table.

	Flowers	Fruits	Honey	Tree exudates	Fleshy fungi	Animals	Insects	Grain	Wood	Milk	Brines	Ocean	Soil	Wine cider
Saccharomyces cerevisiae		+			+	+	+			+			+	+
S. cerevisiae var. *ellipseudes*		+			+								+	
S. rosei							+				+		+	+
Pichia fermentans		+		+		+	+		+				+	
P. membranaefaciens				+		+	+		+		+		+	+
Hansenula anomala		+				+	+	+	+		+			
H. angusta				+		+	+						+	
Debaryomyces klockeri											+		+	+
Cryptococcus albidus	+	+					+			+			+	
Crypto. neoformans		+					+			+				
Crypto. laurentii		+		+			+			+			+	+
Crypto. diffluens		+		+			+			+			+	+
Rhodototula glutinis	+	+		+	+		+		+	+		+	+	
Rh. mucilaginosa	+	+		+		+	+		+	+		+	+	
Rh. minuta				+		+	+			+			+	
Torulopsis colliculosa	+			+	+		+							
T. famata	+	+				+	+		+				+	
T. candida	+			+			+						+	+
T. stellata		+					+							
T. inconspicua				+			+		+				+	
T. glabrata				+		+	+					+		
Torulopsis albida				+			+		+			+	+	
T. aeria				+					+	+		+	+	
Kloeckera magna	+						+				+			
K. apiculata	+	+			+		+						+	+
Candida reukaufii	+	+					+					+		
C. parapsilosis	+	+		+	+	+	+			+			+	
C. guilliermondii	+	+		+		+	+						+	
C. utilis	+					+	+			+				
C. krusei	+	+		+	+	+	+	+		+	+			
C. tropicalis		+				+		+		+			+	
C. zeylanoides		+							+	+			+	
C. mycoderma				+		+	+			+			+	+
C. catenulata				+			+		+					
C. mesenterica				+	+		+						+	+

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