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# Incidence of moulds of dairy plant environment on Khoa

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**Abstract**
Air and khoa samples from three organized dairy plants were collected at different production points namely vat section, package section and storage section in Khoa unit and analysed for the incidence of moulds. *Penicillium citrinum* was found to be predominantly present in all the three sections of khoa unit both in air (23.38%) and khoa samples (27.38%) respectively. The other moulds encountered were *Geotrichum candidum*, *Mucor racemosus*, *Aspergillus niger*, *Syncephalastrum racemosum*, *Aspergillus flavus*, *Fusarium oxysporum*, *Rhizopus stolonifer*, *Cladosporium cladosporioides*, *Absidia corymbifera* and *Paecilomyces variotti*.

**Keywords:** Moulds, dairy plant, khoa unit, vat section, package section, storage section

## 1. Introduction
Khoa is an indigenous dairy product, consumed by large section of population. In India, about 6,00,000 tonnes of khoa is being annually produced by utilizing about 7% of the total milk produced [1]. Khoa serves as a base material for a variety of sweets such as burfi, peda, gulabjamun, kalakand etc. Conventionally, it is prepared by continuous boiling of milk in an open kettle until desired concentration, normally 65-72 per cent total solids and texture is achieved. It is prepared under variety of conditions and gets frequently contaminated by the moulds. The contamination of khoa by moulds causes rapid spoilage of khoa [2]. The presence of moulds in khoa is objectionable as they produce discolouration defects and lipolytic changes causing off flavour development in the finished product [3]. The temperature of khoa at finishing stage is more than 85ºC. It is the post manufacturing contamination that is responsible for high mould count and poor shelf life. Hence the present study was contemplated to establish the incidence of different types of moulds present both in air and khoa samples collected from different sections of khoa making units viz., Vat section, Package section and Storage section and the findings of study will pave way for establishing Good Manufacturing Practices (GMP) in Khoa plant.

## 2. Materials and Methods
### 2.1 Collection of air samples
108 samples of air, 36 from each of the three organized khoa manufacturing dairy plants were collected using Anderson two stage air sampler [4, 5]. Among 36 air samples, 12 samples were collected from each of the three sections in Khoa manufacturing unit *viz.*, Vat section, Package section and Storage section.

### 2.2 Collection of khoa samples
Similarly 108 samples of khoa, 36 from each of the three organized dairies were collected aseptically and analyzed in the laboratory. Among 36 khoa samples, 12 samples each were collected from Vat section, Package section and one week old refrigerated khoa samples from Storage section.

### 2.3 Analysis of moulds in air and khoa samples
The aseptically collected khoa and air samples were analyzed for mould count according to BIS and Andersen [6, 4] respectively. Czapek Dox agar [7] and Lactophenol cotton blue staining technique [8] were employed to observe the morphological and microscopical characteristics of mould isolates.

## 3. Results and Discussion

The mould species of *Aspergilli* were identified as per the procedure of Raper and Fennel [9] and *Pencillium* was identified as per the method of Raper *et al.* [10]. Other genera of fungi were identified by following the keys given by Tandon and Domsch *et al.* [11, 12] and Onions *et al.* [13]. Morphological observations, growth rate, colour of colony, mycelial growth, texture of the culture on agar surface and microscopic appearance in Czapek Dox agar slants, microscopic appearance of petri dishes containing mould growth and micrometric measurement of reproductive structure of lacto phenol cotton blue stained slides for conidiophores and sporangiophores were the important keys followed in identification of mould species.

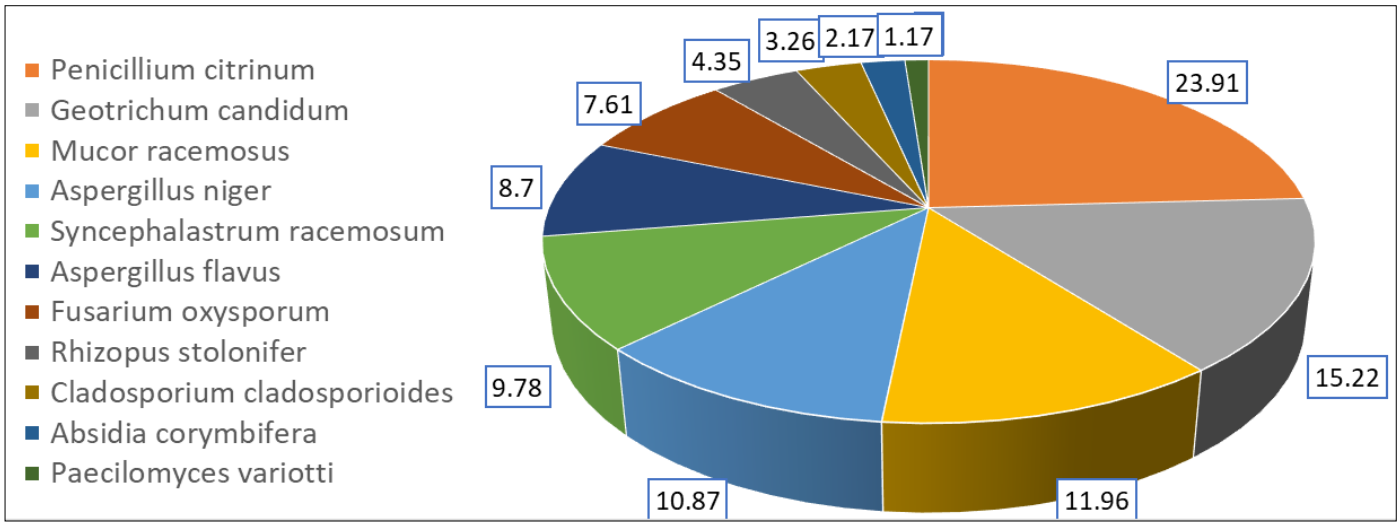


**Fig 1:** Distribution of moulds in Air samples (%)

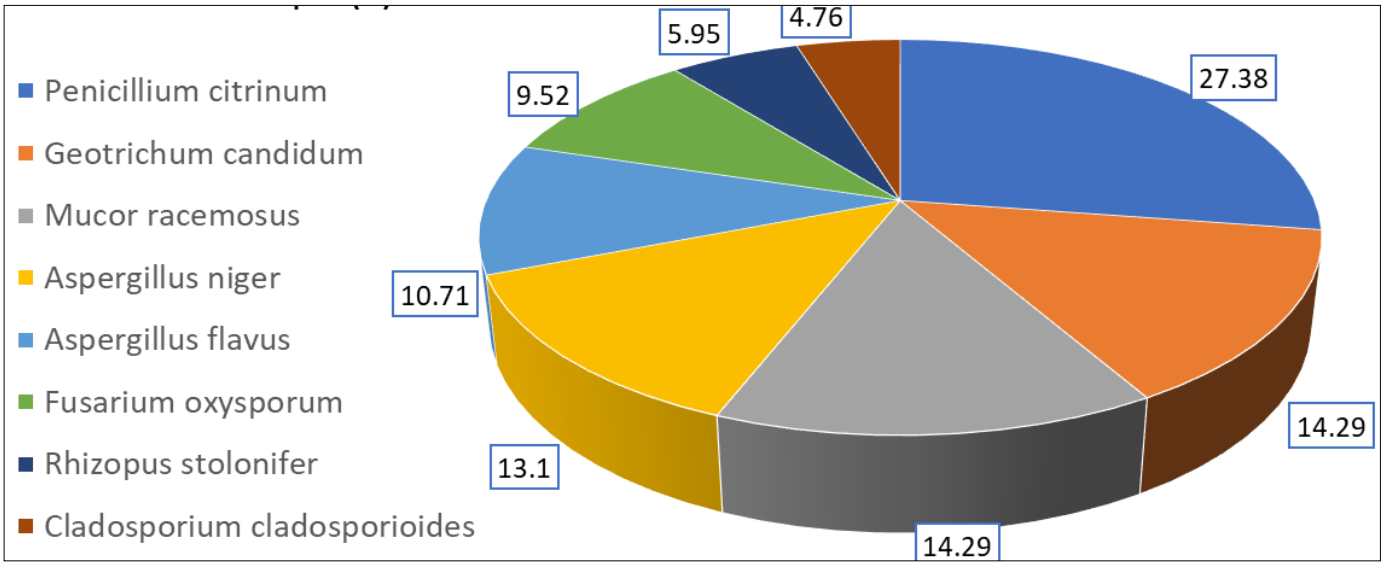


**Fig 2:** Distribution of moulds in Khoa samples (%)

**Table 1:** Distributions of Moulds in Air and Khoa Samples

| S. No | Mould identified | Number isolates | Percentage | Sample sources | | |
|---|---|---|---|---|---|---|
| | | | | Vat section | Package section | Storage section |
| A | In Air Samples | | | | | |
| 1. | *Penicillium citrinum* | 22 | 23.91 | 12 | 7 | 3 |
| 2. | *Geotrichum candidum* | 14 | 15.22 | 7 | 5 | 2 |
| 3. | *Mucor racemosus* | 11 | 11.96 | 6 | 4 | 1 |
| 4. | *Aspergillus niger* | 10 | 10.87 | 6 | 3 | 1 |
| 5. | *Syncephalastrum racemosum* | 9 | 9.78 | 5 | 3 | 1 |
| 6. | *Aspergillus flavus* | 8 | 8.70 | 5 | 2 | 1 |
| 7. | *Fusarium oxysporum* | 7 | 7.61 | 4 | 2 | 1 |
| 8. | *Rhizopus stolonifer* | 4 | 4.35 | 2 | 1 | 1 |
| 9. | *Cladosporium cladosporioides* | 3 | 3.26 | 1 | 1 | 1 |
| 10. | *Absidia corymbifera* | 2 | 2.17 | 2 | Nil | Nil |
| 11. | *Paecilomyces variotti* | 2 | 1.17 | 1 | 1 | Nil |
| | Total | 92 | 100.00 | 51 | 29 | 12 |
| B | In Khoa Samples | | | | | |

| | | | | | | |
|---|---|---|---|---|---|---|
| 1. | *Penicillium citrinum* | 23 | 27.38 | 3 | 8 | 12 |
| 2. | *Geotrichum candidum* | 12 | 14.29 | 2 | 4 | 6 |
| 3. | *Mucor racemosus* | 12 | 14.29 | 1 | 5 | 6 |
| 4. | *Aspergillus niger* | 11 | 13.10 | 1 | 4 | 6 |
| 5. | *Aspergillus flavus* | 9 | 10.71 | 1 | 2 | 5 |
| 6. | *Fusarium oxysporum* | 8 | 9.52 | 1 | 2 | 5 |
| 7. | *Rhizopus stolonifer* | 5 | 5.95 | 1 | 1 | 3 |
| 8. | *Cladosporium cladosporioides* | 4 | 4.76 | 1 | 1 | 2 |
| | Total | 84 | 100.00 | 11 | 28 | 45 |

Table 1 represents the distribution of moulds in air and khoa samples. Out of 92 isolates of air samples, 2 were *Aspergillus flavus*, 10 were *Aspergillus niger*, 3 were *Cladosporium cladosporiodes*, 7 *were Fusarium oxysporum*, 14 were *Geotrichum candidum*, 11 were *Mucor racemosus*, 2 were *Paecilomyces variotti*, 22 were *Penicillium citrinum*, 4 were *Rhizopus stolonifer* and 9 were *Syncephalastrum racemosum*.
Out of 84 isolates of khoa samples, 9 were *Aspergilus flavus*, 11 were *Aspergillus niger*, 4 were *Cladosporium cladosporioides*, 8 were *Fusarium oxysporum*, 12 were *Geotrichum candidum*, 12 were *Mucor racemosus*, 23 were *Penicillium citrinum* and 5 were *Rhizopus stolonifer*.
*Penicillium sp*. (23.91%) and *Aspergillus* sp. (19.57) were the predominant moulds identified in air samples. This is in accordance with the findings of Aggarwal and Srinivasan [14] and Yadav *et al.* [15]. It may be attributed to the prevailing tropical climate and high humidity, poor ventilation systems and outside dust.
Similarly, *Penicillium* sp. (27.38%) and *Aspergillus sp*. (23.81%) were the predominant moulds encountered in khoa samples also. This correlates well with the findings of Yadav *et al*. and Nareshkumar *et al.* [15, 16]. This may be due to faulty handling practices, air movement and improperly cleaned equipments.

## 4. Conclusion
Khoa serves as a favourable medium for the growth of microbes on account of its nutritive value and moisture content. The unsatisfactory condition followed in its production, handling and storage results in poor shelf life. Further, the rapid spoilage of khoa is attributed to its contamination with moulds from environment.
Hence it is concluded that Good Manufacturing Practices (GMP) at all stages of production, packaging and storage of khoa can play a vital role in extending the shelf life of khoa.Proper ventilation in khoa unit along with low humidity and incorporation of permitted antifungal agents namely sorbic acid or benzoic acid may result in production of hygienic khoa with better shelf life.

## 5. Acknowledgement
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