Saccharomycopsis crataegensis,  
a new heterothallic yeast

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A new species, Saccharomycopsis crataegensis, is described. The species is  
heterothallic and forms ellipsoidal ascospores with a single median longitudinal  
ledge. Strains of the species were isolated from grapes and hawthorne fruit  
obtained in Peoria, Illinois U.S.A. A comparison was made between S. cratae-  
gensis and S. vini, which included new strains of the latter species from haw-  
thorne fruit.

INTRODUCTION

A number of strains of Saccharomycopsis vini (Kreger-van Rij) van der Walt  
et Scott were isolated from hawthorne fruit, but among them were four that did  
not sporulate. These isolates were generally similar in colony morphology to  
those of S. vini although they produced only about half as much radial growth.  
When appropriate pairs were mixed, asci formed that contained elongated  
ascospores with a median longitudinal ledge. The carbon assimilation and fer-  
mentation reactions of these strains were similar to S. vini, but it became appar-  
etent that these four isolates, and one obtained earlier from Concord grapes,  
represented a new heterothallic species.

The genus name Endomycopsis Dekker was shown to be illegitimate by van  
der Walt and Scott (1971) since the older genus name Saccharomycopsis  
Schiönnning was also based on the type species of that genus, i.e., E. capsularis  
Schiönnning) Dekker. Because of the heterogeneity of species included in En-  
domycopsis by Kreger-van Rij (1970) and van der Walt and Scott (1971) trans-  
ferred only part of them to Saccharomycopsis and suggested that the remain-  
der be assigned to other genera. Subsequently, von Arx (1972) examined  
these remaining species and assigned them to several other genera. In view of
these revisions, the new species described in this paper was placed in the genus Saccharomycopsis.

MATERIALS AND METHODS

Methods for carrying out fermentation and assimilation tests, and procedures for morphological examination are those of Wickerham (1951). Single-spore cultures of S. vini, used for comparison, were obtained by micromanipulation. Mating tests between S. vini and the mating types of S. crataegensis were carried out as follows: Ascospores of S. vini were freed by digestion with Glusulase¹ (Endo Laboratories, Inc., Garden City, New York) for 15 min at 25 C and streaked on either yeast-malt (YM) agar (Wickerham, 1951) or on restricted growth (RG) agar (Herman, 1971). After 15 to 16 hr, the spores were swollen and beginning vegetative outgrowth. Cells of the mating types of S. crataegensis grown for 15 to 16 hr on YM agar at 25 C were mixed with the germinating spores. Mixtures on RG agar were transferred to YM agar after 24 hr. The mixtures were observed microscopically at 8- to 16-hr intervals for the first 2 days, then at daily intervals for the next 5 days.

Nuclear staining was by the methods of Robinow (1961) but substituting Newcomer's fixative (Newcomer, 1953). A Giemsa stock solution was prepared by adding 0.5 g of stain to 33 ml of glycerol, heating this mixture at 55 C for 2 hr and then adding 33 ml of methanol. The Giemsa stock was diluted 1:20 with 0.1 M phosphate buffer at pH 7.0 just before use.

Ascospores were prepared for scanning electron microscopy (SEM) by first freeing them from the asci by digestion with Glusulase. After digestion for 10–15 min at 25 C, about 50–75% of the spores were freed. Five milliliters of cold (5 C) distilled water was added to the enzyme-cell mixture, and the cells were then removed by centrifugation. The cells were washed three additional times in this manner, and the final pellet was resuspended in 0.2 ml distilled water. Glass squares (8 × 8 mm) cut from microscope slides were cemented to aluminum SEM specimen stages, and a 2-mm loopful of the final cell suspension was spread over the surface of the glass and allowed to air dry at 25 C. The preparations were then vacuum-coated with gold-palladium alloy and viewed in a Cambridge Stereoscan Mark II scanning electron microscope.

¹ Mention of firm names or trade products does not imply that they are endorsed or recommended by the Department of Agriculture over other firms or similar products not mentioned.