

The opinion in support of the decision being entered today was not written for publication and is not binding precedent of the Board.

Paper No. 19

UNITED STATES PATENT AND TRADEMARK OFFICE

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BEFORE THE BOARD OF PATENT APPEALS  
AND INTERFERENCES

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Ex parte LINDA P. THORNE, THOMAS J. POLLOCK  
and RICHARD W. ARMENTROUT

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Appeal No. 1997-2080  
Application No. 08/159,939<sup>1</sup>

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ON BRIEF

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Before WINTERS, SCHEINER and MILLS, Administrative Patent Judges.

SCHEINER, Administrative Patent Judge.

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<sup>1</sup> Application for patent, filed November 30, 1993. According to appellants, this application is a continuation of application serial no. 07/841,716, filed February 26, 1992, now abandoned, which is a divisional of application serial no. 07/777,151, filed October 16, 1991, now U.S. Patent 5,268,460.

DECISION ON APPEAL

This is an appeal under 35 U.S.C. § 134 from the final rejection of claims 7, 21 through 24, and 29 through 35. The final rejection of claims 36 and 37, also pending in the application, was not carried through to the Examiner's Answer, thus claims 36 and 37 are not the subject of appeal.

Claims 21, 29, 31, 33, and 34 are representative of the subject matter on appeal and read as follows:

21. A method for producing substantially non-pigmented pullulan having an  $M_w$  of at least  $6 \times 10^6$  comprising the steps of subjecting a pure culture strain of Aureobasidium pullulans which produces substantially non-pigmented pullulan obtained by

- a) collecting at least one wild-type strain of Aureobasidium pullulans from natural sources;
- b) subjecting the collected strain to fermentation conditions so as to enrich for organisms that grow as fungal yeast-like cells and separating the yeast like cells thus produced;
- c) growing colonies from the separated yeast-like cells;
- d) selecting colonies by visual inspection from step c) which contain yeast-like cells and exhibit reduced pigmentation compared to the other cells;
- e) testing the selected colonies for pigment production; and
- f) selecting the isolate which produces minimum pigment in submerged culture, to fermentation conditions in a fermentation broth and harvesting substantially non-pigmented pullulan having an  $M_w$  of at least  $6 \times 10^6$  from the broth.

29. A method for producing substantially non-pigmented pullulan comprising the steps of subjecting a pure culture strain of Aureobasidium pullulans which produces substantially non-pigmented pullulan obtained by

- a) collecting at least one wild-type strain of Aureobasidium pullulans from natural sources;
- b) subjecting the collected strain to fermentation conditions so as to enrich for organisms that grow as fungal yeast-like cells and separating the yeast like cells thus produced;
- c) growing colonies from the separated yeast-like cells;

- d) selecting colonies by visual inspection from step c) which contain yeast-like cells and exhibit reduced pigmentation compared to the other cells;
- e) testing the selected colonies for pigment production; and
- f) selecting the isolate which produces minimum pigment in submerged culture to fermentation conditions in a fermentation broth and harvesting the substantially non-pigmented pullulan from the broth; and then
- g) subjecting the harvested pullulan to a heat treatment for a period of time and temperature effective to deactivate any enzymes therein which decrease the molecular weight of the pullulan.

31. A method for producing substantially non-pigmented pullulan having a  $M_w$  of at least  $6 \times 10^6$  comprising the steps of subjecting an isolated substantially pure microbial culture selected from the group consisting of those having the following identifying characteristics: A. pullulans ATCC 74100, A. pullulans ATCC 74101, A. pullulans 74102, A. pullulans 74103, A. pullulans 74104, and A. pullulans 74105, and mutants thereof capable of producing substantially non-pigmented pullulan having a  $M_w$  of at least  $6 \times 10^6$ , said cultures being capable of reproducing themselves and of producing pullulan in isolatable amounts when cultured in a liquid growing medium containing assimilable sources of carbon, nitrogen, and inorganic substances to fermentation conditions in a fermentation broth and collecting the substantially non-pigmented pullulan having a molecular weight of at least  $6 \times 10^6$  from the broth.

33. A method for producing substantially non-pigmented pullulan comprising the steps of subjecting a pure culture strain of Aureobasidium pullulans which produces minimal pigment when cultured, obtained by:

- a) collecting at least one wild-type strain of Aureobasidium pullulans from natural sources;
- b) subjecting the collected strain to fermentation conditions so as to enrich for organisms that grow as fungal yeast-like cells;
- c) separating the yeast like cells thus produced;
- d) growing colonies from the separated yeast-like cells;
- e) selecting colonies by visual inspection from step d) which contain yeast-like cells and exhibit reduced pigmentation compared to the other cells;
- f) testing the selected colonies for pigment production; and
- g) selecting the isolate which produces minimum pigment in submerged culture, to fermentation conditions in a fermentation broth, adjusting the pH of the fermentation broth to about 7.0 after the pH of the fermentation broth has stabilized, and thereafter, harvesting the substantially non-pigmented pullulan from the broth; and then subjecting the harvested



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Claims 21, 22, 24, 29 through 32 and 35 stand rejected under 35 U.S.C. 103 as being unpatentable over Shumin Na and Kelly, while claims 7, 23, 33, and 34 stand rejected 35 U.S.C. 103 as being unpatentable over Shumin Na, Kelly and Kato.

We reverse the examiner's rejections.

#### Background

Pullulan, an exopolysaccharide secreted by the fungus Aureobasidium pullulans, is used in transparent films; oxygen-impermeable, tasteless, odorless coatings for food; viscosity control agents; etc. Specification, page 1. A characteristic, and undesirable, property of the fungus is that it produces a melanin-like pigment, dark green to black in color, which contaminates the pullulan during recovery of the product, necessitating a multi-step decoloration treatment before the pullulan can be used. An additional undesirable property is that the average molecular weight of accumulated pullulan decreases as submerged culture in fermentation broth progresses. Id., page 2.

Aureobasidium pullulans is commonly isolated from forest litter, natural waters, wood, leather, plant surfaces, etc. It is polymorphic, exhibiting three predominant forms: elongated branched septate filaments (hyphae); large chlamydospores; and smaller, elliptical yeast-like single cells. Id., pages 1 and 2. According to appellants, "the pigment production characteristic of A. pullulans is associated mainly with the filamentous form and chlamydospores," rather than the yeast-like form. Moreover, wild-type strains "can be treated to enrich the fraction of yeast-like cells therein, and the yeast-like cells can be

separated from the non-yeast-like cells . . . grown and . . . visually isolated based on their degree of pigmentation.” Finally, “pullulan of very high molecular weight may be obtained by subjecting A. pullulans to fermentation conditions and neutralizing the fermentation broth to a pH value of about 7 after the pH broth has stabilized at its characteristic low pH” or by “heat treating the accumulated pullulan for a time period and at a temperature sufficient to deactivate any pullulan decomposing enzymes.” Id., pages 4 and 5.

#### Discussion

In deciding patentability issues under 35 U.S.C. § 103, the court observed in Panduit Corp. v. Dennison Mfg. Co., 810 F.2d 1561, 1567-68, 1 USPQ2d 1593, 1597 (Fed. Cir. 1987), “[a]nalysis begins with a key legal question -- what is the invention claimed?” since “[c]laim interpretation . . . will normally control the remainder of the decisional process.”

All of the present claims are directed to a method of producing substantially non-pigmented pullulan by subjecting A. pullulans to a fermentation process. As maintained by appellants, and as acknowledged by the examiner, certain of the claims additionally require enrichment of A. pullulans cultures for yeast-like cells, followed by selection of a non-pigmented strain (claims 7, 21-24, 29, 30, 33, and 36). Brief, page 12 and Examiner’s Answer, page 3. Claims 31, 32, 34, 35, and 37 require the use of specific strains of A. pullulans (or mutants thereof with defined properties); claims 24, 29, 30, 33, 34, 36 and 37 require post-harvest heating of the pullulan to inactivate enzymes; claims 7,

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21-24, 30-32 and 35 require production and recovery of pullulan of a specific minimum molecular weight; and claims 7, 23, 33, 34, 36 and 37 impose a pH limitation upon the fermentation conditions. In summary, each of the claims requires one or more of the following: enrichment of A. pullulans cultures for yeast-like cells, followed by selection of a non-pigmented strain; specific strains of A. pullulans; heat treatment of harvested pullulan to inactivate enzymes; production and recovery of pullulan of defined molecular weight; and particular pH conditions.

#### Rejection I

According to the examiner, “[Shumin Na] disclose[s] culturing strain[s] of Aureobasidium pullulans which were milky white” and “also teach[es] that non-pigmented strain A22 had yeast characteristics and resulted in high pullulan yield” and “unpigmented extracellular polysaccharide can be obtained from it.” Further according to the examiner, [Kelly] disclose[s] a process of enriching for organism[s] that grow as fungal yeast-like cells by treating Aureobasidium pullulans with ethidium bromide [] and subjecting pullulan to heat treatment.” Examiner’s Answer, page 3.

The examiner concludes that the invention of claims 21, 22, 24, 29-32 and 35 is unpatentable over Shumin Na and Kelly because “a person having ordinary skill in the art . . . would have been motivated to enrich [Shumin Na’s] yeast-like cells of strain A22 . . . and to select yeast-like cells which exhibit reduced pigmentation in order to produce non-pigmented pullulans.”

We disagree. As set forth in In re Kotzab, 217 F.3d 1365, 1369-70, 55 USPQ2d 1313, 1316 (Fed. Cir. 2000):

A critical step in analyzing the patentability of claims pursuant to section 103(a) is casting the mind back to the time of invention, to consider the thinking of one of ordinary skill in the art, guided only by the prior art references and the then-accepted wisdom in the field. [] Close adherence to this methodology is especially important in cases where the very ease with which the invention can be understood may prompt one “to fall victim to the insidious effect of a hindsight syndrome wherein that which only the invention taught is used against its teacher.” [citations omitted]

. . . [T]o establish obviousness based on a combination of the elements disclosed in the prior art, there must be some motivation, suggestion or teaching of the desirability of making the specific combination that was made by the applicant.

The examiner’s rejection rests on the assertion that one would have been motivated to enrich the proportion of yeast-like cells in a culture of A22 and to select cells exhibiting reduced pigmentation in order to produce non-pigmented pullulans. This statement of the rejection is, quite simply, untenable on its face. Shumin Na states on page 5 that “[t]he fermentation solutions of strain A22 . . . were always milky white” and “[t]he silver mirror test indicated there existed only yeast type cells, no chlamyospores or arthrospores were produced.” Since A22 is already yeast-like and already produces nonpigmented pullulan, we see nothing in Shumin Na or Kelly which would provide a reason to subject the culture to enrichment or further selection.

Moreover, we see nothing in Shumin Na or Kelly which would suggest producing non-pigmented pullulan by subjecting other strains of A. pullulans to the specific series of

enrichment and selection steps required by the claims. Shumin Na isolated thirty-six strains of Aureobasidium from cocoons of the Canadian leaf-cutting insect. Those strains that produced an extracellular polysaccharide, produced non-pigmented pullulan -- in contrast to a reference strain, AS 3,3984, which produced darkly pigmented pullulan. When the “cell morphology of AS 3,3984 and the relatively high polysaccharide-yield strain A22 [were compared,] the results indicated that both strains had yeast and hypha morphological characteristics.” Shumin Na, page 5. The authors conclude that “Strain A22 is different than the pigmented varieties of Aureobasidium isolated . . . in the tropics and subtropics.” Id., page 8. In our view, if anything, Shumin Na would suggest that pigment production in Aureobasidium varies from strain to strain, rather than morphological form. Kelly adds little or nothing to Shumin Na’s teachings, as the reference merely teaches that “there is accumulating evidence that the elaboration of pullulan is associated with the yeast-like phase of A. pullulans,” but does not indicate that pullulan produced by yeast-like cells is unpigmented.

Finally, the statement of the rejection does not begin to address the various additional requirements of certain of the claims: e.g., recovery of pullulan of  $M_w$   $6 \times 10^6$  or greater; heat treatment of harvested pullulan to inactivate degradative enzymes; and/or the use of particular strains of A. pullulans. 35 U.S.C. § 103 requires that obviousness be determined based on the claimed subject matter as a whole. Where, as here, the

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determination of obviousness was based on less than the entire claimed subject matter, the examiner's conclusion of obviousness is legally unsound and cannot stand.

On this record, we reverse the examiner's rejection of claims 21, 22, 24, 29-32 and 35 under 35 U.S.C. § 103.

### Rejection II

Claims 7, 23, 33, and 34 require adjusting the pH of the fermentation broth to about 7.0 after the pH of the fermentation broth has stabilized, and stand rejected under 35 U.S.C. § 103 as being unpatentable over Shumin Na, Kelly and Kato. The examiner relies on Kato to establish that "lowering the initial pH of the culture medium greatly increases the molecular weight of pullulans obtained." Examiner's Answer, pages 3 and 4. From this, the examiner concludes that "in order to prepare a pullulan of high molecular weight, a person having ordinary skill in the art . . . would have been motivated to start fermentation at a lower pH and adjust the pH to neutral after the initial fermentation has taken place." Id., page 4.

We disagree. We find no suggestion in Kato to adjust the pH to neutral after the initial fermentation has taken place and the pH has stabilized. Moreover, Kato does nothing to overcome the underlying deficiency in the examiner's proposed combination of the teachings of Shumin Na and Kelly.

Accordingly, we reverse the examiner's rejection of claims 7, 23, 33 and 34 under 35 U.S.C. § 103 as well.

CONCLUSION

For the reasons set forth in the body of this opinion, we reverse the rejection of claims 21, 22, 24, 29-32 and 35 as unpatentable over Shumin Na and Kelly; as well as the rejection of claims 7, 23, 33 and 34 as unpatentable over Shumin Na, Kelly and Kato.

REVERSED

	)	
Sherman D. Winters	)	
Administrative Patent Judge	)	
	)	
	)	
	)	BOARD OF PATENT
Toni R. Scheiner	)	
Administrative Patent Judge	)	APPEALS AND
	)	
	)	INTERFERENCES
	)	
Demetra J. Mills	)	
Administrative Patent Judge	)	

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