UDC 663.12:579.234:547.96
ISSN 1330–9862

review

Saccharomyces cerevisiae Cell Wall Proteins*

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Received: December 7, 1998
Accepted: February 15, 1999

Summary

Cell wall of Saccharomyces cerevisiae contains more than 20 different mannoproteins. They are considered to play different roles in building, maintaining and modifying the wall itself when different cell cycle events, or other conditions, require so. Besides, they are important for interactions of cells with their surrounding, the example of which are intercellular interactions during agglutination or flocculation. Cell wall proteins can be divided in three groups according to the mechanism by which they are attached to the wall. Some proteins are connected to wall structural polysaccharides noncovalently and the identification of several members of this group showed that they most probably all possess enzymatic activities. The second group comprises proteins covalently attached, most probably to β-1,6-glucan, and they can be released from the wall by different glucanase preparations. Finally, a group of proteins can be extracted from the wall by 30 mM NaOH but the actual link by which they are attached to the cell wall is unknown.

In this paper a survey of so far identified and partially characterised cell wall proteins of each of the three groups is presented. Besides, the present knowledge of how the proteins get incorporated into the wall as well as their structure and potential role is summarised.

Key words: yeast cell wall, mannoproteins, cell wall biosynthesis, glucanases

Introduction

Yeast cell wall represents a vital extracellular organelle which provides the necessary mechanical stability of the cell, but is also involved in the communication of the cell with its surrounding, fulfilling in this way a number of important cellular requirements. Even though the outstanding features of this structure withstanding turgor pressures larger than 15 bar (*1*), and still maintaining significant flexibility necessary for the cell growth and budding, have been noticed long ago, the cell wall nevertheless represents one of the most neglected cellular organelles. In contrast to bacterial cell walls, the structure of the yeast wall is still not well understood. Little is known about biochemical mechanisms concerning the synthesis of individual components of the cell wall and particularly about ways how these components are interconnected into a rigid network. The reason lies, at least partly, in the relative complexity of the yeast cell wall. It is composed of two major components, β-1,3-glucan and mannoproteins, the former building the inner layer, and the latter forming the outer layer of the wall (*2–4*). Besides, the cell wall contains two minor, but not less important components, β-1,6-glucan, supposed to serve as a link between mannoproteins and β-1,3-glucan (*5,6*), and chitin, which is predominantly located in rings formed between mother and daughter cell during budding (*7,8*), but is to some extent also distributed throughout the whole cell wall (*9,10*). While the carbohydrate components of the cell wall, in particular

* Dedicated to Professor Pavao Mildner for his 80th birdthday

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β-1,3-glucan, have a role in the mechanical stabilisation of the cell, it is not clear at all what the functions of the cell wall mannoproteins are, most of which can be removed from the wall without any change in shape or osmotic stability of the cell. It has been assumed that some of the cell wall proteins serve in building up the wall itself and are required in addition for structural changes in the wall during particular cell cycle events like budding, mating, or sporulation. Others may be required for interactions of the yeast cell with its surrounding, as is the case in cell-cell interactions in the processes of agglutination (*11,12*) and flocculation (*13*). Systematic analysis of *Saccharomyces cerevisiae* cell wall proteins showed that they could be divided in two groups. Some proteins were released from the cell wall by heating it in SDS and mercaptoethanol, while others withstood this treatment and were extracted from the wall only by enzymatic degradation of glucan using different glucanase preparations like zymolyase (*14*) or laminarinase (*15*). The first group of proteins was assumed noncovalently attached to carbohydrate components of the wall, while the latter was thought to comprise proteins covalently anchored in the wall. A significant problem in attempts to identify cell wall proteins, however, occurred due to glucan attracting proteins nonspecifically, particularly at acidic pH (*16*). As a result, upon mechanical disruption of cells, cell walls were heavily contaminated with intracellular proteins. Therefore, N-terminal sequencing of first isolated »cell wall proteins« revealed sequences of glycolytic enzymes (R. Teparić, C. B. Sharma, unpublished results). Actually, the existence of enolase in *Candida* cell walls was reported (*17*). To overcome this problem, a method for specific labeling of cell wall proteins by biotinylation was introduced (*18,19*). Using this method, followed by the visualisation of labeled proteins after electrophoresis and blotting by the streptavidin-peroxydase conjugate, 10 SDS-soluble and 7 glucanase-soluble proteins were identified. Besides, it has been shown that some proteins which remain in the cell wall after the hot SDS/mercaptoethanol treatment were released by an overnight incubation in 30 mM NaOH, thus indicating that there are at least three different ways of building proteins into the cell wall (*19*).

The aim of this review is to summarise our present knowledge about these three groups of S. *cerevisiae* cell wall proteins. It will particularly be discussed how these proteins are embedded into the wall, their structural properties and potential roles in this cellular structure.

Noncovalently Linked Cell Wall Proteins

Generally, proteins noncovalently attached to structural polysaccharides of the cell wall have been extracted using 1–2 % SDS solution containing up to 5 % β-mercaptoethanol, or often using the Laemmli sample electrophoresis buffer. Extraction was usually performed 5–10 minutes at 95–100 °C (*14,15,20*). Alternatively, a very similar protein pattern has been obtained when cell walls were treated with 2 mM DTT overnight at 4 °C (*20*), indicating that these proteins are disulfide-bound to other proteins, possibly to proteins covalently linked to cell wall components.

Little is known about the sorting mechanism responsible for the attachment of cell wall proteins to wall polysaccharides, since no apparent structural properties distinguish these proteins from those secreted into the medium, or those retained by the wall as soluble periplasmic proteins. It has been shown that, due to a high hydrogen bonding potential, β-1,3-glucan can react with many proteins *in vitro* at acidic pH (*16*). The exceptions were, as expected, periplasmic enzymes invertase and acid phosphatase. It could also be demonstrated that mechanical disruption of cells at acidic pH result in interactions of a number of intracellular proteins with cell walls, while the addition of these proteins to whole cells did not bring about such interactions, indicating that they were specific for the internal, glucan layer of the cell wall (*21*). Therefore, it may be speculated that no particular mechanism is required for the embedding of proteins, but that rather those proteins not retained by the wall share some common physico-chemical property.

The first two noncovalently attached cell wall proteins described, chitinase (Cts1p; *22*) and an exoglucanase (Exg1p; *23*), were actually identified in the growth medium. A more precise study of the localization of these two enzymes revealed that about 10 % of each protein could be found in the cell wall as well. As a matter of fact, it is assumed that the wall fraction of chitinase is the one with physiological significance, since its substrate is wall chitin itself. Cts1p is the enzyme responsible for the release of the daughter from the mother cell by lysing the chitin ring after the completion of the primary and the secondary septum between the two cells (*22*). The protein contains a C-terminal domain which binds with high affinity specifically to chitin and provides the required concentration of the enzyme in the region of the bud neck. Since the role and the physiological substrate of the Exg1p is not known, the importance of the cell wall fraction of this protein cannot be assessed.

The first integral cell wall protein which was purified, and whose gene was cloned and sequenced was the Bgl2p protein (*24*). Biochemical characterisation of this protein first suggested that it was a β-1,3-exoglucanase, but more precise studies showed that it rather exhibited β-1,3-endoglucanase (*25*), or transglucosidase (*26*) activities *in vitro*, depending on the concentration of the substrate. Since the disruption of *BGL2* did not bring about any apparent phenotype (*24*), the role of the protein in the cell wall is still unknown.

Labeling of cell wall proteins by biotinylation proved to be a most useful method for their identification and characterisation. Extraction of labeled proteins with hot SDS/mercaptoethanol led to the identification of 9 protein bands upon blotting of the extracted proteins (*19*). Visualised proteins were named Scw1p-Scw9p (Soluble cell wall proteins). Since the reaction of Scw proteins with SDS made their subsequent purification difficult, alternative extraction procedures were tested. It was found that the incubation of cell walls in 2 mM DTT resulted in the release of a very similar pattern of proteins (*20*), which were then purified and N-terminally sequenced. This strategy led to the identification of 7 most prominent Scw proteins. Among the sequenced proteins were also the three previously characterised cell wall proteins, namely Scw2p was found to be identical

to Cts1p (chitinase), Scw6p was identical to Exg1p, and Scw9p to Bgl2p. This demonstrated the specificity of the labeling procedure used and the fact that none of the intracellular proteins were labeled suggested that previous findings of glycolytic enzymes associated with the cell wall (*17*) were most probably artifacts.

Purification of biotinylated DTT-soluble proteins revealed three new, so far unidentified wall proteins designated as Scw3p, Scw4p and Scw10p; the latter comigrated with Scw4p in electrophoresis. Scw4p and Scw10p represented the most prominent band among Scws and their purification and identification by N-terminal sequencing revealed that the two proteins were indeed of the same size and highly homologous, sharing 63 % sequence identity. Comparison with other *Sacch. cerevisiae* proteins, as well as with other available data libraries at the DNA and protein level revealed that Scw4p and Scw10p were homologous to several glucanases or glucanase-related proteins. Interestingly, the highest homology was observed with the Bgl2p (*20*). Therefore, the two Scw proteins were purified and a potential hydrolytic activity was tested *in vitro* using different polysaccharide substrates like laminarin, yeast β-1,3-glucan, chitin, pustulan, p-nitrophenylglucoside, etc. Neither of the proteins hydrolysed any of these substrates indicating that they probably catalyse a more complex reaction in the wall. Single disruptions of either *SCW4* or *SCW10* did not bring about any apparent phenotype, but a double mutant was shown to be more susceptible to cell wall synthesis inhibitors Calcofluor white and Congo red. Besides, the double mutant possessed a decreased mating ability. Cell walls of the *scw4 scw10* mutant contained significantly less glucanase-soluble proteins indicating that the two proteins might play a role, directly or indirectly, in the covalent attachment of proteins to glucan (*20*).

SCW3 was compared with genes available in different data banks. Interestingly, the highest homology was found between *SCW3* and the *Candida wickerhamii* gene coding for a β-glucanase which was thought to play a nutritional role when this organism was grown on cellodextrins as a carbon source (*27*). It is not known if Scw3p plays a similar role in *Sacch. cerevisiae*.

Finally, Scw8p was also identified and it was found to be identical to Ccw5p, a protein covalently attached in the cell wall (see later). Therefore, it can be assumed that Scw8p represents either an intermediate, or an improperly localised form of Ccw5p (*20*).

Summarising the data available on proteins noncovalently attached or S-S-linked in the *Sacch. cerevisiae* cell wall, it can be concluded that they all either represent enzymes with demonstrated hydrolytic, in particular glycolytic activities, or that they are homologous to such enzymes. It has to be mentioned, however, that they do not necessarily all have to represent hydrolases but that some of them could also act as transglycosidases, in that way contributing to the overall structure and rigidity of the cell wall. It can be speculated that the flexibility of the wall is achieved by a counterplay of different glycosidases and transglycosidases, which could also explain a relatively high number of these enzymes in the cell wall. Noncovalently linked cell wall proteins identified so far are listed in Table 1.

Table 1. Noncovalently linked cell wall proteins

Protein	YPD-code	Size*	Function	Ref.
Cts1p/Scw2p	YLR286c	116	chitinase	*20, 22*
Scw3p	YNL066w	95	glucanase homologue	*20*
Scw4p	YGR279c	66	glucanase homologue	*20*
Scw10p	YMR305c	66	glucanase homologue	*20*
Exg1p/Scw6p	YLR300w	44	exoglucanase	*20, 23*
Scw8p/Ccw5p	YJL158c	41	?	*20*
Bgl2p/Scw9p	YGR282c	29	endoglucanase/ transglucosidase	*20, 24–26*

* as relative molecular mass / kDa

Covalently Linked Cell Wall Proteins Extractable by Glucanases

Extensive washing of yeast cell walls with reagents like SDS, mercaptoethanol, urea, guanidinium hydrochloride, or DTT, even at increased temperatures, cannot deplete all proteins. Based on this fact, the remaining proteins are considered covalently attached to the structural polysaccharide components of the cell wall. Valentin *et al.* (*14*) have first shown that some proteins can be released from the purified, hot SDS/mercaptoethanol treated cell walls using zymolyase, a glucanase preparation. Subsequent investigations showed that laminarinase, another commercial preparation used for the lysis of yeast cell walls, was more appropriate for obtaining distinct protein bands (*15,19*).

The first protein of this class studied in more detail was α-agglutinin (*28,29*), a mating type specific protein which is, upon pheromone induction, secreted into the cell wall where it promotes agglutination reacting specifically with a-agglutinin, its counterpart in cells of the opposite mating type (*30,31*). It has been found that the α-agglutinin contains a putative GPI-anchoring signal at its C-terminus and the study of intermediate forms of the molecule throughout the secretory pathway, on its way to the cell wall, indicated that this protein transverses the whole route in a GPI-anchored form. Upon its localisation in the plasma membrane, it is released by splitting one of the linkages within the GPI-anchor and is translocated to the cell wall as shown in Fig.1. (*32*).

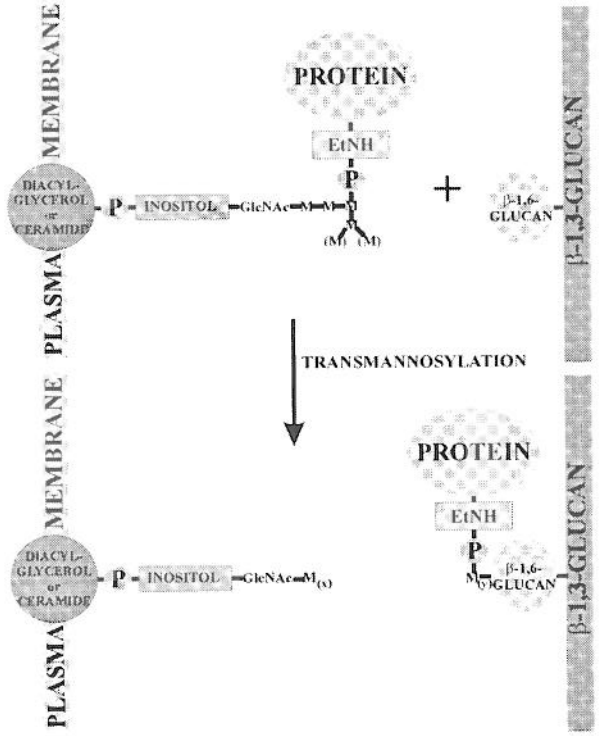

Fig. 1. Schematic presentation of the tentative GPI-anchoring mechanism of glucanase-extractable covalently linked cell wall proteins

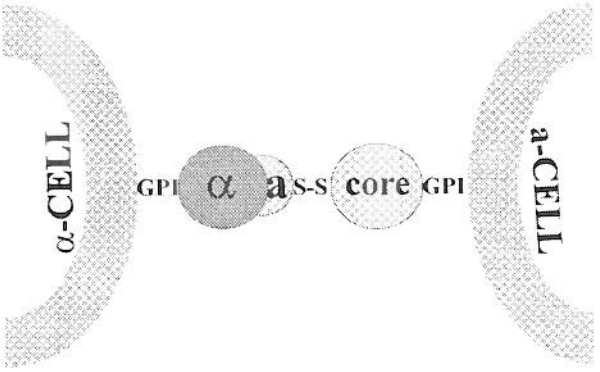

Fig. 2. Intermolecular interactions of proteins involved in agglutination

Identification of three more glucanase-extractable wall proteins, named Cwp1p, Cwp2p and Tip1p, showed that they also contained potential GPI-anchoring signals (*33*), suggesting that this way of attaching proteins to the cell wall represents a general principle, rather than being specific for α-agglutinin. At the same time it has been postulated that the recipient polysaccharide component of the wall might be β-1,6-glucan, since the proteins released by laminarinase still reacted with anti-β-1,6-glucane antibodies (*15,33*). It was postulated that the transfer of a GPI-anchored protein to β-1,6-glucan involves a transmannosylation reaction (*6,34*), but details of this mechanism are still unknown.

Besides the four proteins mentioned, another seven glucanase-extractable cell wall proteins have been purified and identified so far. Aga1p was characterised as a part of the a-agglutinin complex (Fig.2.) consisting of the a-agglutinin protein (Aga2p) which interacts directly with α-agglutinin in the agglutination process, and the Aga1p, a so called »core« protein which is anchored in the wall by a GPI-mediated mechanism (*35*). The two proteins are linked by two disulfide bonds (*31*) and no other role of the Aga1p except of anchoring the a-agglutinin in the wall is known. Although β-mercaptoethanol and DTT enhance the release of noncovalently attached proteins from the wall, for none of these proteins a link to some other wall protein through a disulfide bridge has actually been demonstrated. Therefore, the attachment of a-agglutinin remains the only example so far for such localisation mechanism in *Sacch. cerevisiae*.

Another function for a yeast cell wall mannoprotein is flocculation. This asexual reversible aggregation of cells is generally thought to involve lectin-type interactions of particular wall proteins with mannan moieties. Three genes, *FLO1, FLO5* and *FLO8* have been genetically defined to be required for flocculation (*36*), and Flo1p has been purified from laminarinase cell wall extracts (*37*). Even though a substantial amount of this protein was also released by a prolonged and repeated hot SDS-treatment, it is still likely that it is predominantly bound to the cell wall by the GPI-anchor mediated mechanism. This was supported by the fact that a deletion of the C-terminal domain of the protein, containing the putative GPI-anchoring signal, led to the secretion of the remaining polypeptide into the medium. It also resulted in the loss of the flocculation ability, indicating that Flo1p is the factor directly responsible for intercellular interactions during flocculation (*37*).

Icwp1p was characterised via epitope towards a monoclonal antibody raised to *Sacch. cerevisiae* cell wall extract (*38*). Immunofluorescence analysis showed that the protein was located at the inner layer of the cell wall and that it was only accessible to antibodies if the mannoprotein layer was affected by tunicamycin. The possible function of this protein is unknown, but it was found that the disruption of the corresponding gene led to an increased sensitivity of cells to zymolyase, and to cell wall synthesis inhibitors Calcofluor white and Congo red. The authors proposed a structural role for this protein (*38*).

Sed1p has been extracted from purified cell walls by a digestion with the *Rarobacter faecitabidus* protease I, a serine proteinase which recognises mannan chains and cleaves peptide bonds in their vicinity. The protein was characterised as one of the major cell wall proteins in stationary cells (*39*). Interestingly, the *SED1* gene was first characterised as a multy-copy suppresor of *erd1* which was deficient in retaining ER-proteins (*40*). A link between this process and the overproduction of a cell wall protein, however, is unclear. The only apparent phenotype of the *SED1* disruption is an increased sensitivity of cells to zymolyase.

Another protein with relative molecular mass of about 100 kDa has been purified using the same protease from cell walls of static culture cells, but it was absent in cells grown with aeration (*41*). This protein was iden-

tified as a product of the *TIR1/SRP1* gene which had previously been characterised as a gene induced by glucose, cold shock or anaerobiosis. It was also found that its transcription was regulated by the Rox1p repressor (*41*). The extracted protein reacted with anti-β-1,6-glucan antibodies indicating that it was bound to this wall polysaccharide, similarly as other cell wall proteins of this class.

Labeling of *S. cerevisiae* surface proteins by biotinylation and their systematic identification revealed most of the covalently linked cell wall proteins described in other studies. Five bands of biotinylated proteins were visualised after electrophoresis and blotting (*19*). Additionally, two proteins were identified in the stacking gel of the electrophoretic system (V. Mrša *et al.*, unpublished results). The first five bands have been designated Ccw1p--Ccw5p, while the latter two were named Ccw12p and Ccw13p. N-terminal sequencing of proteins isolated from the gel revealed that Ccw1p was identical to Icwp1p, Ccw2p was Tip1p, and Ccw3p was Tip1p plus Cwp1p (M. Ecker, unpublished results). Ccw4p was not identified so far, while Ccw5p was a new, so far unidentified cell wall protein which, however, possessed all structural characteristics of a third class of wall proteins, extractable with NaOH. Since this protein was indeed also found in NaOH extracts of the wall, it will be discussed below. Ccw12p was identified as a product of the previously described *α0.6* gene which is negatively regulated by the mating pheromone α-factor (*42*), while Ccw13p was shown to be a product of the *DAN1* gene, which is induced by the anaerobic growth conditions (*43*). Physiological roles of these proteins are unknown.

In conclusion, the class of glucanase-extractable cell wall proteins comprises mannoproteins which share a great deal of structural similarity. They all possess putative GPI-anchoring signals at their C-termini required for their proper localisation. Besides, they are all rich in hydroxy amino acids which are often clustered in particular parts of their sequences. Finally, they are all abundantly glycosylated, both through N-glycosylation, as well as by O-glycosylation. Little is known about the physiological role of this group of proteins with the exception of those induced during the particular events like agglutination or flocculation. The mutations of some of them were reported to lead to the increased sensitivity and weakening of the cell wall (*38,39*, V. Mrša *et al.*, unpublished results). However, a multiple mutant lacking *CCW12, CCW13, CWP1, TIP1* and *ICWP1* genes (thus lacking all of the most prominent electrophoretic bands of this class of proteins) still did not show any significant growth problem, nor did it require osmotic stabilisation (M. Ecker, unpublished results). Interestingly, it seems that perhaps a whole set of cell wall proteins is induced during anaerobic growth of cells, although a connection between anaerobiosis and cell wall structure and properties is not at all clear. All proteins of this class identified so far are summarised in Table 2.

Covalently linked cell wall proteins extractable by NaOH

A particular class of cell wall proteins which has recently been discovered (*19*) comprise a set of mannoproteins extractable by an overnight incubation of purified,

Table 2. Covalently linked cell wall proteins

Protein	YPD-code	Extractable by	Size*	GPI-signal	Function/ regulation	Ref.
Ccw12p/α0.6p	YLR110c	glucanases	>300	YES	down-regulated by α-factor	*42*, V. Mrša *et al.*, unpublished
Ccw13p/Dan1p	YJR150c	glucanases	>300	YES	anaerobic growth	*43*, V. Mrša *et al.*, unpublished
Flo1p	YAR050w	glucanases	>300	YES	flocculation	*37*
Sed1p	YDR077w	*R. faecitabidus* proteinase I, glucanases	300	YES	stationary cells	*39*
Agα1p	YJR004c	glucanases	250	YES	α-agglutinin	*28, 29, 30*
Aga1p	YNR044c	glucanases	73	YES	»core« part of a-agglutinin	*35*
Tir1p/Srp1p	YER011w	*R. faecitabidus* proteinase I, glucanases	100	YES	anaerobic growth	*41*
Cwp1p	YKL096w	glucanases	55	YES	?	*33*
Tip1p	YBR067c	glucanases	80	YES	?	*33*
Cwp2p	YKL444	glucanases	180	YES	?	*33*
Icwp1p/Ccw1p	YLR391w	glucanases	250	YES	?	*38*, V. Mrša *et al.*, unpublished
Ccw5p/Ccw11p/ Scw8p	YJL158c	glucanases, NaOH	41	NO	?	*19*
Ccw6p/Pir1p	YKL164c	NaOH	250	NO	?	*19*
Ccw7p	YJL159c	NaOH	115	NO	?	*19*
Ccw8p/Pir3p	YKL163w	NaOH	57	NO	?	*19*

* as relative molecular mass / kDa

SDS extracted cell walls in 30 mM NaOH at 4 °C (Table 2.). Six protein bands have been revealed upon biotinylation of surface proteins and they were named Ccw6p-Ccw11p. N-terminal sequencing of Ccw11p, however, showed that it was the same as Ccw5p, a protein already identified in the laminarinase wall extracts. Therefore, this designation will be further used for this protein. Besides Ccw5p, three other proteins of this class, Ccw6p, Ccw7p and Ccw8p have been identified by N-terminal sequencing. *CCW6* gene was found to be identical to the previously described *PIR1*, while *CCW8* was the same as *PIR3*. These two genes were discovered in a study involving sequencing of the gene *FSR2* adjacent to *PIR1*. Southern blot crosshybridization revealed another two homologous genes *PIR2* and *PIR3*. The gene family originally attracted attention due to the relatively long repetitive sequence of its members. Pir2p has in the meantime been identified as a heat shock induced protein named HSP150, found to be secreted into the medium (*44*). The proteins Pir1p and Pir3p have been located in the cell wall and labeled by biotinylation (*19*). N-terminal sequencing of Ccw5p and Ccw7p revealed that these two proteins also belong to the same protein family sharing a high degree of homology and several common structural features. All of them contain the same characteristic repetitive sequence at their N-terminal end, the number of repeating units ranging between 2 (Ccw5p) and 10 (Ccw7p). Further, they all contain the recognition site for the Kex2p proteinase, and their isolation from *kex2* mutant showed that in all of them this site was really recognised and processed (*19*). Finally, none of the proteins of this family contain the C-terminal GPI-anchoring signal characteristic for other covalently attached cell wall proteins. The fact that they remain associated with the wall even after extensive treatment with hot SDS/mercaptoethanol (except of the Ccw5p which is released to a minor extent) suggests that this protein family is covalently linked to wall structural polysaccharides. However, the absence of the GPI-anchoring signal, and the fact that these proteins (except Ccw5p) do not seem to be released by glucanases, indicate that the way of their attachment and the chemical link is different from that characteristic for proteins released by laminarinase. Since they are extracted from the wall under mild β-elimination conditions, and since it has been found that all of them are O-glycosylated to a high degree, it can be speculated that the O-glycosidic mannan chains are somehow involved in their attachment in the cell wall.

Nothing is known about the possible function of this protein family in the cell wall. Disruption of individual genes encoding its members did not bring about any apparent phenotype, as expected due to the high similarity of these proteins (*19*). A disruption of *CCW6/PIR1*, however, only in combination with the *PIR2/HSP150* was reported to lead to a decreased thermal stability of the mutant (*45*). Members of the family located in the cell wall alone were shown not to contribute to this property, nor were they regulated by a heat shock, as it has been found for *HSP150* (*19*). Therefore, a multiple mutation of different, and maybe all members of this protein family is required to enlighten their physiological role.

Conclusion

A number of studies on yeast cell wall proteins, predominantly performed with *Saccharomyces cerevisiae*, showed that there are at least three classes of proteins which differ in their chemical links as well as in the way by which they get attached to this extracellular organelle. Analysis of physiological functions of these proteins indicate that their way of anchoring may reflect their role in the wall. So, enzymes required for the formation, maintenance and changes in the structure of the wall are found among the proteins noncovalently attached. Covalently linked proteins are often considered structural components, although a relatively high number and variability among these proteins suggest that they could also have other, so far unknown functions. Proteins which were extracted from the wall by NaOH are still the least studied, but the fact that they all belong to the same protein family indicate that their function in this cellular structure is also somehow related to the way of their localisation. Further work is required to identify remaining cell wall proteins, elucidate molecular mechanism involved in their targeting and anchoring in the wall and, finally, to learn more about their physiological function in this outermost cellular compartment.

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Stanična stijenka kvasca *Saccharomyces cerevisiae*

Sažetak

Stanična stijenka kvasca Saccharomyces cerevisiae sadrži više od 20 različitih manoproteina. Drži se da oni imaju različite uloge u izgradnji, održavanju i promjenama u samoj staničnoj stijenci kada to zahtijevaju pojedini događaji u staničnom ciklusu ili drugi uvjeti. Osim toga, oni su važni za interakcije stanice sa svojom okolinom, kao npr. interakcije između stanica tijekom aglutinacije ili flokulacije. Proteini stanične stijenke mogu se prema mehanizmu kojim su pričvršćeni u stijenci podijeliti u tri skupine. Neki su proteini nekovalentno povezani sa strukturnim polisaharidima stijenke, a identifikacija nekolicine članova te skupine pokazala je da svi oni vjerojatno imaju enzimsku aktivnost. Druga skupina obuhvaća kovalentno vezane proteine, vjerojatno na β-1,6-glukan i oni se mogu osloboditi iz stijenke različitim preparacijama glukanaza. Konačno, jedna se skupina proteina može ekstrahirati iz staničnih stijenki pomoću 30 mM NaOH, ali nije poznata veza kojom su oni povezani sa stijenkom.

U ovom je radu dan pregled do danas identificiranih i djelomično karakteriziranih proteina stanične stijenke svake od navedenih skupina. Osim toga, sažeto je prikazano kako se proteini ugrađuju u stijenku, te opisana njihova struktura i uloga.