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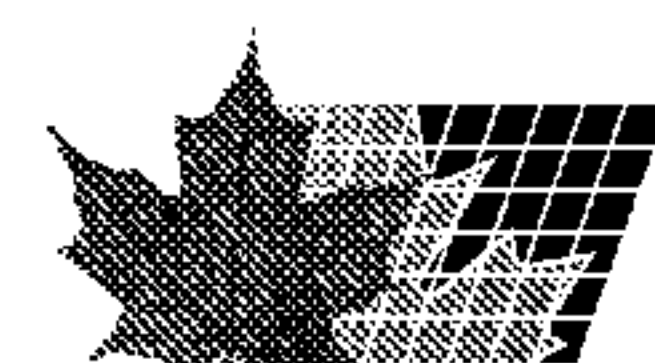
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(54) Titre : PROCEDE DE GENERATION D'UNE RESISTANCE A DES AGENTS PATHOGENES DANS DES PLANTES

(54) Title: METHOD FOR OBTAINING THE PATHOGENIC RESISTANCE IN PLANTS

(57) **Abrégé/Abstract:**

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(54) Title: METHOD FOR OBTAINING THE PATHOGENIC RESISTANCE IN PLANTS

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METHOD FOR OBTAINING THE PATHOGENIC RESISTANCE IN PLANTS

The invention relates to methods for generating or increasing a pathogen resistance in plants by reducing the expression, activity or function of an NADPH oxidase.

10 The aim of plant biotechnology work is the generation of plants with advantageous novel properties, for example for increasing agricultural productivity. The plants' natural defense mechanisms against pathogens are frequently insufficient. Fungal diseases alone result in annual yield losses of many billions of US\$. The introduction of foreign genes from plants, animals or microbial sources can increase the defenses. Examples are the protection against feeding damage by insects by expressing *Bacillus thuringiensis* endotoxins (Vaeck et al. (1987) Nature 328:33-37) or the protection against fungal infection by expressing a bean chitinase (Broglie et al. (1991) Science 254:1194-1197). However, most of the approaches described only offer resistance to a single pathogen or a narrow spectrum of pathogens.

20 Only a few approaches exist which impart a resistance to a broader spectrum of pathogens to plants. Systemic acquired resistance (SAR) - a defense mechanism in a variety of plant/pathogen interactions - can be conferred by the application of endogenous messenger substances such as jasmonic acid (JA) or salicylic acid (SA) (Ward, et al. (1991) Plant Cell 3:1085-1094; Uknes, et al. (1992) Plant Cell 4(6):645-656). Similar effects can also be achieved by synthetic compounds such as 2,6-dichloroisonicotinic acid (INA) or S-methyl benzo(1,2,3)thiadiazole-7-thiocarboxylate (BTH; Bion®) (Friedrich et al. (1996) Plant J 10(1):61-70; Lawton et al. (1996) Plant J. 10:71-82). The expression of pathogenesis-related (PR) proteins, which are upregulated in the case of SAR, may also cause pathogen resistance in some cases.

30 In barley, the Mlo locus has been described as a negative regulator of the defense against pathogens. The loss of the Mlo gene causes an increased and, above all, race-unspecific resistance against a large number of mildew species (Büschges R et al. (1997) Cell 88:695-705; Jorgensen JH (1977) Euphytica 26:55-62; Lyngkjaer MF et al. (1995) Plant Pathol 44:786-790). Mlo-deficient barley varieties obtained by conventional breeding are already being used in agriculture. Despite intensive cultivation, the resistance has proved to be durable, presumably due to the fact that it is recessive. Mlo-like resistances in other plants,

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in particular in cereal species, are not described. The Mlo gene and various homologs from other cereal species have been identified and cloned (Büschges R et al. (1997) Cell 88:695-705; WO 98/04586; Schulze-Lefert P, Vogel J (2000) Trends Plant Sci. 5:343-348). Various methods using these genes for obtaining a pathogen resistance are described (WO 98/04586; WO 00/01722; WO 99/47552). The disadvantage is that the Mlo-mediated defense mechanism comprises a spontaneous die-off of leaf cells (Wolter M et al. (1993) Mol Gen Genet 239:122-128). Another disadvantage is that the Mlo-deficient genotypes show hypersensitivity to hemi-biotrophic pathogens such as *Magnaporthe grisea* (*M. grisea*) and *Cochliobolus sativus* (*Bipolaris sorokiniana*) (Jarosch B et al. (1999) Mol Plant Microbe Interact 12:508-514; Kumar J et al. (2001) Phytopathology 91:127-133).

15 The liberation of reactive oxygen species (ROS; for example superoxide (O_2^-), hydroxyl radicals and H_2O_2) is ascribed an important protection function in the reaction on plant pathogens (Wojtaszek P (1997) Biochem J 322:681-692). A variety of ways of how a cell can produce ROS are known. In the macrophages of mammals, it is in particular the enzyme NADPH oxidase, which is able to transfer electrons to molecular oxygen, which must be mentioned. Homologous enzymes have also been identified in plants (Lamb & Dixon (1997) Annu Rev Plant Physiol Plant Mol Biol 48:251).

25 It has been shown that mutations in the catalytic subunit of NADPH oxidase in *Arabidopsis thaliana* show a reduced accumulation of reactive oxygen intermediates (ROI). With regard to the hypersensitive reaction (HR), the results were heterogeneous: while infection with the avirulent and bacterium *Pseudomonas syringae* showed a reduced HR in a double mutant, the virulent oomycete *Peronospora parasitica* showed an increased HR. Growth - both of virulent and of avirulent *P. syringae* strains - was not changed in comparison with wild-type plants, however (Torres MA et al. (2002) Proc Natl Acad Sci USA 99:517-522). Likewise, the inhibition of NADPH oxidase by means of the inhibitor diphenyleneiodonium chloride (DPI) - at physiologically relevant concentrations - had no effect on the development of pathogenic fungi (Hückelhoven R & Kogel KH (1998) Mol Plant Microbe Interact 11:292-300). A cDNA fragment of a phagocytic barley NADPH oxidase (pNAox, homolog of the large subunit *gp91phox* of a phagocytic NADPH oxidase) is described under the GenBank Acc.-No.: AJ251717).

The present invention aims at providing novel compounds for the defense against pathogens in plants, which compounds bring about an efficient defense against as broad as possible a pathogen spectrum in as many different plant species as possible, prefer-

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ably the crop plants used in agriculture. We have found that this object is achieved by the present method.

A first aspect of the invention comprises a method for generating or increasing the resistance to at least one pathogen in plants, which comprises the following operating steps:

- a) reduction of the protein quantity, activity or function of an NADPH oxidase in a plant or a tissue, organ, part or cell thereof, and
- b) selection of the plants in which - in contrast or in comparison with the starting plant - the resistance to at least one pathogen exists or is increased.

15

Surprisingly, the reduction of the expression of a barley NADPH oxidase (pNAox) in the epidermal cell by a sequence-specific RNA interference approach using double-stranded pNAox-dsRNA ("gene silencing") shows a significantly reduced disease level following Bgh infection (measured with reference to the formulation of Haustoria). This finding is particularly surprising because the release of reactive oxygen species ("oxidative burst"), which is associated with NADPH oxidase, is generally ascribed a protective function.

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Similar to Mlo, the reduction of the NADPH oxidase expression mediates a broad resistance to various isolates of *Blumeria graminis* f.sp. *hordei*. In transient gene silencing experiments, the penetration efficiency (development of Haustoria) of Bgh is reduced significantly by more than 35% - an effect which, in its intensity, corresponds to the effect obtained by means of Mlo-dsRNA (Schweizer P et al. (2000) Plant J 24:895-903). In the wild-type barley variety Pallas, approximately 40% of the fungal penetrations result in the development of haustoria, while the penetration rate in the case of reduced NADPH oxidase expression by introduction of a double-stranded RNA of NADPH oxidase (pNAox-dsRNA) only amounts to approximately 25%. The fact that even in pathogen-sensitive wild-type varieties such as Pallas only a penetration rate of approximately 40 to 50% can be observed can be attributed to the basal resistance, which is always present. Owing to these findings, the enzyme NADPH oxidase can be considered a key element for the successful penetration of a pathogen such as Bgh into the plant cell. In addition, the method is superior to all those methods where a pathogen-resistant phenotype is generated by overexpression of a resistance-mediating protein. Switching off a gene can be done without expression of a (foreign) protein. In the ideal case, only the endogenous gene is de-

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activated. This has not inconsiderable advantages regarding approval and acceptance by the consumer, who is frequently unsure about plants with foreign proteins. Very especially advantageous in this context is the use of inducible promoters for reducing
5 the NADPH oxidase quantity, activity or function, which, for example in the case of pathogen-inducible promoters, makes possible an expression only when required (i.e. attack by pathogens).

In principle, the method according to the invention can be applied
10 plied to all plant species, preferably to those in which an NADPH oxidase or a functional equivalent thereof is expressed naturally.

For the purposes of the invention, "plant" means all genera and
15 species of higher and lower plants of the Plant Kingdom. Included in this expression are the mature plants, seed, shoots and seedlings, and parts, propagation material, plant organs, tissues, protoplasts, callus and other cultures, for example cell cultures, derived from them, and all other types of groups of plant
20 cells which give functional or structural units. Mature plants refers to plants at any developmental stage beyond that of the seedling. Seedling means a young, immature plant in an early developmental stage. "Plant" comprises all annual and perennial monocotyledonous and dicotyledonous plants and includes by way of
25 example, but not by limitation, those of the genera Cucurbita, Rosa, Vitis, Juglans, Fragaria, Lotus, Medicago, Onobrychis, Trifolium, Trigonella, Vigna, Citrus, Linum, Geranium, Manihot, Daucus, Arabidopsis, Brassica, Raphanus, Sinapis, Atropa, Capsicum, Datura, Hyoscyamus, Lycopersicon, Nicotiana, Solarium, Petunia,
30 Digitalis, Majorana, Cichorium, Helianthus, Lactuca, Bromus, Asparagus, Antirrhinum, Heterocallis, Nemesis, Pelargonium, Panicleum, Pennisetum, Ranunculus, Senecio, Salpiglossis, Cucumis, Browaalia, Glycine, Pisum, Phaseolus, Lolium, Oryza, Zea, Avena, Hordeum, Secale, Triticum, Sorghum, Picea and Populus.

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The term "plant" preferably comprises monocotyledonous crop plants, such as, for example, cereal species such as wheat, barley, millet, rye, triticale, maize, rice, sorghum or oats, and sugar cane.

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The term furthermore comprises dicotyledonous crop plants such as, for example

- Brassicaceae such as oilseed rape, canola, cress, Arabidopsis,
45 sis, cabbages or canola, Leguminosae such as soybean, alfalfa, pea, beans or peanut

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- Solanaceae such as potato, tobacco, tomato, egg plant or paprika, Asteraceae such as sunflower, Tagetes, lettuce or Calendula,
- 5 - Cucurbitaceae such as melon, pumpkin/squash or zucchini, and linseed, cotton, hemp, clover, spinach, flax, red pepper, carrot, beet, radish, sugar beet, sweet potato, cucumber, chichory, cauliflower, broccoli, asparagus, onion, garlic, celeriac,
- 10 strawberry, raspberry, blackberry, pineapple, avocado, and the various tree, bush, nut and vine species. Tree species preferably comprises plum, cherry, peach, nectarine, apricot, banana, paw paw, mango, apple, pear, quince.
- 15 Furthermore comprised are ornamental plants, useful or ornamental trees, flowers, cut flowers, shrubs or lawn, by way of example but not by way of limitation, the families of the Rosaceae such as rose, Ericaceae such as rhododendrons and azaleas, Euphorbiaceae such as poinsettias and croton, Caryophyllaceae such as
- 20 carnations, Solanaceae such as petunias, Gesneriaceae such as African violets, Balsaminaceae such as touch-me-not, Orchidaceae such as orchids, Iridaceae such as gladioli, iris, freesia and crocus, Compositae such as calendula, Geraniaceae such as geraniums, Liliaceae such as dracaena, Moraceae such as ficus, Araceae
- 25 such as philodendron, and many others.

Preferred for the purposes of the invention are those plants which are employed as food or feedstuff, very especially preferably monocotyledonous genera and species, such as the above-

30 described cereal species.

The method is very especially preferably applied to monocotyledonous plants, most preferably to plants with agricultural importance such as wheat, oats, millet, barley, rye, maize, rice,

35 buckwheat, sorghum, triticale, spelt, linseed or sugar cane.

"Pathogen resistance" denotes the reduction or weakening of disease symptoms of a plant following infection by a pathogen. The symptoms can be manifold, but preferably comprise those which

40 directly or indirectly have an adverse effect on the quality of the plant, the quantity of the yield, the suitability for use as feedstuff or foodstuff, or else which make sowing, planting, harvesting or processing of the crop difficult.

45 "Conferring", "existing", "generating" or "increasing" a pathogen resistance means that the defense mechanisms of a specific plant species or variety is increasingly resistant to one or more

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pathogens due to the use of the method according to the invention in comparison with the wild type of the plant ("original plant"), to which the method according to the invention has not been applied, under otherwise identical conditions (such as, for
5 example, climatic conditions, growing conditions, pathogen species and the like). The increased resistance manifests itself preferably in a reduced manifestation of the disease symptoms, disease symptoms comprising - in addition to the abovementioned adverse effects - for example also the penetration efficiency of
10 a pathogen into the plant or plant cells or the proliferation efficiency in or on the same. In this context, the disease symptoms are preferably reduced by at least 10% or at least 20%, especially preferably by at least 40% or 60%, very especially preferably by at least 70% or 80% and most preferably by at least
15 90% or 95%.

"Selection" with regard to plants in which - as opposed or as compared to the original plant - resistance to at least one pathogen exists or is increased means all those methods which are
20 suitable for recognizing an existing or increased resistance to pathogens. These may be symptoms of pathogen infection (for example the development of haustoria in the case of fungal infection), but may also comprise the above-described symptoms which relate to the quality of the plant, the quantity of the
25 yield, the suitability for use as feedstuff or foodstuff and the like.

"Pathogen" within the scope of the invention means by way of example but not by limitation viruses or viroids, bacteria,
30 fungi, animal pests such as, for example, insects or nematodes. Especially preferred are fungi, such as mildew. However, it can be assumed that the expression of an NADPH oxidase, its activity or its function also brings about resistance to other pathogens. The following pathogens may be mentioned by way of example but
35 not by limitation:

1. Fungal pathogens and fungus-like pathogens:

Fungal pathogens and fungus-like pathogens (such as, for
40 example, Chromista) are preferably from the group comprising Plasmodiophoromycota, Oomycota, Ascomycota, Chytridiomycetes, Zygomycetes, Basidiomycota and Deuteromycetes (Fungi imperfecti). The pathogens mentioned in Tables 1 and 2 and the diseases with which they are associated may be mentioned
45 by way of example but not by limitation.

Table 1: Fungal plant diseases

	Disease	Pathogen
5	Leaf ruse	Puccinia recondita
	Yellow rust	P. striiformis
	Powdery mildew	Erysiphe graminis / Blumeria graminis
	Glume blotch	Septoria nodorum
10	Leaf blotch	Septoria tritici
	Ear fusarioses	Fusarium spp.
	Eyespot	Pseudocercospora herpotrichoides
	Smut	Ustilago spp.
15	Bunt	Tilletia caries
	Take-all	Gaeumannomyces graminis
	Anthracnose leaf blight	Colletotrichum graminicola (teleomorph: Glomerella graminicola)
	Anthracnose stalk rot	Politis); Glomerella tucumanensis (anamorph: Glomerella falcatum Went)
20	Aspergillus ear and kernel rot	Aspergillus flavus
	Banded leaf and sheath spot	Rhizoctonia solani Kuhn = Rhizoctonia microsclerotia J. Matz (telomorph: Thanatephorus cucumeris)
25	Black bundle disease	Acremonium strictum W. Gams = Cephalosporium acremonium Auct. non Corda
	Black kernel rot	Lasioidiplodia theobromae = Botryodiplodia theobromae
30	Borde blanco	Marasmiellus sp.
	Brown spot (black spot, stalk rot)	Physoderma maydis
	Cephalosporium kernel rot	Acremonium strictum = Cephalosporium acremonium
35	Charcoal rot	Macrophomina phaseolina
	Corticium ear rot	Thanatephorus cucumeris = Corticium sasakii
40	Curvularia leaf spot	Curvularia clavata, C. eragrostidis = C. maculans (teleomorph: Cochliobolus eragrostidis), Curvularia inaequalis, C. intermedia (teleomorph: Cochliobolus intermedius), Curvularia lunata (teleomorph: Cochliobolus lunatus), Curvularia pallescens (teleomorph: Cochliobolus pallescens), Curvularia senegalensis, C. tuberculata (teleomorph: Cochliobolus tuberculatus)
45	Didymella leaf spot	Didymella exitalis

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	Disease	Pathogen
	Diplodia ear rot and stalk rot	Diplodia frumenti (teleomorph: Botryosphaeria festucae)
5	Diplodia ear rot, stalk rot, seed rot and seedling blight	Diplodia maydis = Stenocarpella maydis
	Diplodia leaf spot or streak	Stenocarpella macrospora = Diplodia leaf macrospora

10

Table 2: Downy mildew

	Disease	Pathogen
15	Brown stripe downy mildew	Sclerophthora rayssiae var. zeae
	Crazy top downy mildew	Sclerophthora macrospora = Sclerospora macrospora
20	Green ear downy mildew (graminicola downy mildew)	Sclerospora graminicola
	Java downy mildew	Peronosclerospora maydis = Sclerospora maydis
	Philippine downy mildew	Peronosclerospora philippinensis = Sclerospora philippinensis
25	Sorghum downy mildew	Peronosclerospora sorghi = Sclerospora sorghi
	Spontaneum downy mildew	Peronosclerospora spontanea = Sclerospora spontanea
	Sugarcane downy mildew	Peronosclerospora sacchari = Sclerospora sacchari
30	Dry ear rot (cob, kernel and stalk rot)	Nigrospora oryzae (teleomorph: Khuskia oryzae)
35	Ear rots, minor	Alternaria alternata = A. tenuis, Aspergillus glaucus, A. niger, Aspergillus spp., Botrytis cinerea (teleomorph: Botryotinia fuckeliana), Cunninghamella sp., Curvularia pallescens, Doratomyces stemonitis = Cephalotrichum stemonitis, Fusarium culmorum, Gonatobotrys simplex, Pithomyces maydis, Rhizopus microsporus Tiegh., R. stolonifer = R. nigricans, Scopulariopsis brumptii
40		
45	Ergot (horse's tooth)	Claviceps gigantea (anamorph: Sphacelia sp.)
	Eyespot	Aureobasidium zeae = Kabatiella zeae

	Disease	Pathogen
	Fusarium ear and stalk rot	Fusarium subglutinans = F. moniliforme var. subglutinans
5	Fusarium kernel, root and stalk rot, seed rot and seedling blight	Fusarium moniliforme (teleomorph: Gibberella fujikuroi)
	Fusarium stalk rot, seedling root rot	Fusarium avenaceum (teleomorph: Gibberella avenacea)
10	Gibberella ear and stalk rot	Gibberella zeae (anamorph: Fusarium graminearum)
	Gray ear rot	Botryosphaeria zeae = Physalospora zeae (anamorph: Macrophoma zeae)
	Gray leaf spot (Cercospora leaf spot)	Cercospora sorghi = C. sorghi var. maydis, C. zeae-maydis
15	Helminthosporium root rot	Exserohilum pedicellatum = Helminthosporium pedicellatum (teleomorph: Setosphaeria pedicellata)
	Hormodendrum ear rot (Cladosporium rot)	Cladosporium cladosporioides = Hormodendrum cladosporioides, C. herbarum (teleomorph: Mycosphaerella tassiana)
20	Hyalothyridium leaf spot	Hyalothyridium maydis
	Late wilt	Cephalosporium maydis
25	Leaf spots, minor	Alternaria alternata, Ascochyta maydis, A. tritici, A. zeicola, Bipolaris victoriae = Helminthosporium victoriae (teleomorph: Cochliobolus victoriae), C. sativus (anamorph: Bipolaris sorokiniana = H. sorokinianum = H. sativum), Epicoccum nigrum,
30		Exserohilum prolatum = Drechslera prolata (teleomorph: Setosphaeria prolata)
35		Graphium penicilliioides, Leptosphaeria maydis, Leptothyrium zeae, Ophiosphaerella herpotricha, (anamorph: Scolecosporiella sp.), Paraphaeosphaeria michotii, Phoma sp., Septoria zeae, S. zeicola, S. zeina
40	Northern corn leaf blight (white blast, crown stalk rot, stripe)	Setosphaeria turcica (anamorph: Exserohilum turcicum = Helminthosporium turcicum)
	Northern corn leaf spot Helminthosporium ear rot (race 1)	Cochliobolus carbonum (anamorph: Bipolaris zeicola = Helminthosporium carbonum)
45	Penicillium ear rot (blue eye, blue mold)	Penicillium spp., P. chrysogenum, P. expansum, P. oxalicum
	Phaeocystostroma stalk rot and root rot	Phaeocystostroma ambiguum, = Phaeocystosporella zeae

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	Disease	Pathogen
	Phaeosphaeria leaf spot	Phaeosphaeria maydis = Sphaerulina maydis
5	Physalospora ear rot (Botryosphaeria ear rot)	Botryosphaeria festucae = Physalospora zeicola (anamorph: Diplodia frumenti)
	Purple leaf sheath	Hemiparasitic bacteria and fungi
	Pyrenochaeta stalk rot and root rot	Phoma terrestris = Pyrenochaeta terrestris
10	Pythium root rot	Pythium spp., P. arrhenomanes, P. graminicola
	Pythium stalk rot	Pythium aphanidermatum = P. butleri L.
	Red kernel disease (ear mold, leaf and seed rot)	Epicoccum nigrum
15	Rhizoctonia ear rot (sclerotial rot)	Rhizoctonia zeae (teleomorph: Waitea circinata)
	Rhizoctonia root rot and stalk rot	Rhizoctonia solani, Rhizoctonia zeae
20	Root rots, minor	Alternaria alternata, Cercospora sorghi, Dictyochoeta fertilis, Fusarium acuminatum (teleomorph: Gibberella acuminata), F. equiseti (teleomorph: G. intricans), F. oxysporum, F. pallidoroseum, F. poae, F. roseum, G. cyanogena, (anamorph: F. sulphureum), Microdochium bolleyi, Mucor sp., Periconia circinata, Phytophthora cactorum, P. drechsleri, P. nicotianae var. parasitica, Rhizopus arrhizus
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30	Rostratum leaf spot (Helminthosporium leaf disease, ear and stalk rot)	Setosphaeria rostrata, (anamorph: Exserohilum rostratum = Helminthosporium rostratum)
	Rust, common corn	Puccinia sorghi
	Rust, southern corn	Puccinia polysora
35	Rust, tropical corn	Physopella pallescens, P. zeae = Angiopsora zeae
	Sclerotium ear rot (southern blight)	Sclerotium rolfsii Sacc. (teleomorph: Athelia rolfsii)
40	Seed rot-seedling blight	Bipolaris sorokiniana, B. zeicola = Helminthosporium carbonum, Diplodia maydis, Exserohilum pedicillatum, Exserohilum turcicum = Helminthosporium turcicum, Fusarium avenaceum, F. culmorum, F. moniliforme, Gibberella zeae (anamorph: F. graminearum), Macrophoma phaseolina, Penicillium spp., Phomopsis sp., Pythium spp., Rhizoctonia solani, R. zeae, Sclerotium rolfsii, Spicaria sp.
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	Disease	Pathogen
	Selenophoma leaf spot	Selenophoma sp.
	Sheath rot	Gaeumannomyces graminis
5	Shuck rot	Myrothecium gramineum
	Silage mold	Monascus purpureus, M ruber
	Smut, common	Ustilago zeae = U. maydis
	Smut, false	Ustilaginoidea virens
10	Smut, head	Sphacelotheca reiliana = Sporisorium holcisorghi
	Southern corn leaf blight and stalk rot	Cochliobolus heterostrophus (anamorph: Bipolaris maydis = Helminthosporium maydis)
15	Southern leaf spot	Stenocarpella macrospora = Diplodia macrospora
20	Stalk rots, minor	Cercospora sorghi, Fusarium episphaeria, F. merismoides, F. oxysporum Schlechtend, F. poae, F. roseum, F. solani (teleomorph: Nectria haematococca), F. tricinctum, Mariannaea elegans, Mucor sp., Rhopoglyphus zeae, Spicaria sp.
	Storage rots	Aspergillus spp., Penicillium spp. and other fungi
	Tar spot	Phyllachora maydis
25	Trichoderma ear rot and root rot	Trichoderma viride = T. lignorum teleomorph: Hypocrea sp.
	White ear rot, root and stalk rot	Stenocarpella maydis = Diplodia zeae
30	Yellow leaf blight	Ascochyta ischaemi, Phyllosticta maydis (teleomorph: Mycosphaerella zeae-maydis)
	Zonate leaf spot	Gloeocercospora sorghi

35 The following are especially preferred

- Plasmodiophoromycota such as Plasmodiophora brassicae (clubroot of crucifers), Spongospora subterranea (powdery scab of potato tubers), Polymyxa graminis (root disease of cereals and grasses),
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- Oomycota such as Bremia lactucae (downy mildew of lettuce), Peronospora (downy mildew) in snapdragon (P. antirrhini), onion (P. destructor), spinach (P. effusa), soybean (P. manchurica), tobacco ("blue mold"; P. tabacina), alfalfa and clover (P. trifolium), Pseudoperonospora humuli (downy mildew of hops), Plasmopara (downy mildew in grapevines) (P.
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- viticola) and sunflower (*P. halstedii*), *Sclerophthora macrospora* (downy mildew in cereals and grasses), *Pythium* (seed rot, seedling damping-off, and root rot of all types of plants, for example damping-off of Beta beet caused by *P. debaryanum*), *Phytophthora infestans* (blight in potato, brown rot in tomato and the like), *Albugo spec.* (white rust on cruciferous plants).
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- 10 - Ascomycota such as *Microdochium nivale* (snow mold of rye and wheat), *Fusarium graminearum*, *Fusarium culmorum* (partial ear sterility mainly in wheat), *Fusarium oxysporum* (*Fusarium* wilt of tomato), *Blumeria graminis* (powdery mildew of barley (f.sp. *hordei*) and wheat (f.sp. *tritici*)), *Erysiphe pisi* (powdery mildew of pea), *Nectria galligena* (*Nectria* canker of
- 15 fruit trees), *Uncinula necator* (powdery mildew of grapevine), *Pseudopeziza tracheiphila* (red fire disease of grapevine), *Claviceps purpurea* (ergot on, for example, rye and grasses), *Gaeumannomyces graminis* (take-all on wheat, rye and other grasses), *Magnaporthe grisea* (rice blast disease),
- 20 *Pyrenophora graminea* (leaf stripe of barley), *Pyrenophora teres* (net blotch of barley), *Pyrenophora tritici-repentis* (leaf blight of wheat), *Venturia inaequalis* (apple scab), *Sclerotinia sclerotium* (stalk break, stem rot), *Pseudopeziza medicaginis* (leaf spot of alfalfa, white and red clover).
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- Basidiomycetes such as *Typhula incarnata* (typhula blight on barley, rye, wheat), *Ustilago maydis* (blister smut on maize), *Ustilago nuda* (loose smut on barley), *Ustilago tritici* (loose smut on wheat, spelt), *Ustilago avenae* (loose smut on oats),
- 30 *Rhizoctonia solani* (*rhizoctonia* root rot of potato), *Sphacelotheca* spp. (head smut of sorghum), *Melampsora lini* (rust of flax), *Puccinia graminis* (stem rust of wheat, barley, rye, oats), *Puccinia recondita* (leaf rust on wheat), *Puccinia dispersa* (brown rust on rye), *Puccinia hordei* (leaf
- 35 rust of barley), *Puccinia coronata* (crown rust of oats), *Puccinia striiformis* (yellow rust of wheat, barley, rye and a large number of grasses), *Uromyces appendiculatus* (brown rust of bean), *Sclerotium rolfsii* (root and stem rots of many plants).
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- Deuteromycetes (*Fungi imperfecti*) such as *Septoria nodorum* (glume blotch) of wheat (*Septoria tritici*), *Pseudocercospora herpotrichoides* (eyespot of wheat, barley, rye), *Rhynchosporium secalis* (leaf spot on rye and
- 45 barley), *Alternaria solani* (early blight of potato, tomato), *Phoma betae* (blackleg on Beta beet), *Cercospora beticola* (leaf spot on Beta beet), *Alternaria brassicae* (black spot on

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oilseed rape, cabbage and other crucifers), *Verticillium dahliae* (verticillium wilt), *Colletotrichum lindemuthianum* (bean anthracnose), *Phoma lingam* (blackleg of cabbage and oilseed rape), *Botrytis cinerea* (gray mold of grapevine, strawberry, tomato, hops and the like).

Most preferred are *Phytophthora infestans* (potato blight, brown rot in tomato and the like), *Microdochium nivale* (previously *Fusarium nivale*; snow mold of rye and wheat), *Fusarium* 10 *graminearum*, *Fusarium culmorum* (partial ear sterility of wheat), *Fusarium oxysporum* (*Fusarium* wilt of tomato), *Blumeria graminis* (powdery mildew of barley (f. sp. *hordei*) and wheat (f. sp. *tritici*)), *Magnaporthe grisea* (rice blast disease), *Sclerotinia sclerotium* (stalk break, stem rot), *Septoria nodorum* and *Septoria* 15 *tritici* (glume blotch of wheat), *Alternaria brassicae* (black spot of oilseed rape, cabbage and other crucifers), *Phoma lingam* (blackleg of cabbage and oilseed rape).

2. Bacterial pathogens:

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The pathogens and diseases associated with them, all of which are mentioned in table 3, may be mentioned by way of example, but not by limitation.

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Table 3: Bacterial diseases

	Disease	Pathogen
	Bacterial leaf blight and stalk rot	<i>Pseudomonas avenae</i> subsp. <i>avenae</i>
30	Bacterial leaf spot	<i>Xanthomonas campestris</i> pv. <i>holcicola</i>
	Bacterial stalk rot	<i>Enterobacter dissolvens</i> = <i>Erwinia dissolvens</i>
	Bacterial stalk and top rot	<i>Erwinia carotovora</i> subsp. <i>carotovora</i> , <i>Erwinia chrysanthemi</i> pv. <i>zeae</i>
35	Bacterial stripe	<i>Pseudomonas andropogonis</i>
	Chocolate spot	<i>Pseudomonas syringae</i> pv. <i>coronafaciens</i>
40	Goss's bacterial wilt and blight (leaf freckles and wilt)	<i>Clavibacter michiganensis</i> subsp. <i>nebraskensis</i> = <i>Corynebacterium michiganense</i> pv. <i>nebraskense</i>
	Holcus spot	<i>Pseudomonas syringae</i> pv. <i>syringae</i>
	Purple leaf sheath	Hemiparasitic bacteria
	Seed rot-seedling blight	<i>Bacillus subtilis</i>

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Stewart's disease (bacterial wilt)	Pantoea stewartii = Erwinia stewartii
5 Corn stunt (achapparramiento, maize stunt, Mesa Central or Rio Grande maize stunt)	Spiroplasma kunkelii

Very especially preferred are the following pathogenic bacteria:
 Corynebacterium sepedonicum (potato bacterial ring rot), Erwinia
 10 carotovora (potato bacterial soft rot), Erwinia amylovora (fire
 blight on pear, apple, quince), Streptomyces scabies (potato
 scab), Pseudomonas syringae pv. tabaci (tobacco black fire),
 Pseudomonas syringae pv. phaseolicola (bean grease spot), Pseudo-
 monas syringae pv. tomato (tomato bacterial speck), Xanthomonas
 15 campestris pv. malvacearum (cotton bacterial blight) and Xantho-
 monas campestris pv. oryzae (bacterial leaf blight on rice and
 other grasses).

3. Viral pathogens:

- 20 "Viral pathogens" includes all plant viruses such as, for ex-
 ample, tobacco or cucumber mosaic virus, ringspot virus, ne-
 croses virus, maize dwarf mosaic virus and the like.
- 25 Pathogens and the diseases associated with them may be men-
 tioned in table 4 by way of example, but not by limitation.

Table 4: Viral diseases

30	Disease	Pathogen
	American wheat striate (wheat striate mosaic)	American wheat striate mosaic virus (AWSMV)
	Barley stripe mosaic	Barley stripe mosaic virus (BSMV)
	Barley yellow dwarf	Barley yellow dwarf virus (BYDV)
35	Brome mosaic	Brome mosaic virus (BMV)
	Cereal chlorotic mottle	Cereal chlorotic mottle virus (CCMV)
	Corn chlorotic vein banding (Brazilian maize mosaic)	Corn chlorotic vein banding virus (CCVBV)
40	Corn lethal necrosis	Virus complex of Maize chlorotic mottle virus (MCMV) and Maize dwarf mosaic virus (MDMV) A or B or Wheat streak mosaic virus (WSMV)
	Cucumber mosaic	Cucumber mosaic virus (CMV)
45	Cynodon chlorotic streak	Cynodon chlorotic streak virus (CCSV)
	Johnsongrass mosaic	Johnsongrass mosaic virus (JGMV)

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	Disease	Pathogen
	Maize bushy stunt	Mycoplasma-like organism (MLO) associated
5	Maize chlorotic dwarf	Maize chlorotic dwarf virus (MCDV)
	Maize chlorotic mottle	Maize chlorotic mottle virus (MCMV)
	Maize dwarf mosaic	Maize dwarf mosaic virus (MDMV) strains A, D, E and F
	Maize leaf fleck	Maize leaf fleck virus (MLFV)
10	Maize line	Maize line virus (MLV)
	Maize mosaic (corn leaf stripe, enanismo rayado)	Maize mosaic virus (MMV)
	Maize mottle and chlorotic stunt	Maize mottle and chlorotic stunt virus
15	Maize pellucid ringspot	Maize pellucid ringspot virus (MPRV)
	Maize raya gruesa	Maize raya gruesa virus (MRGV)
	maize rayado fino (fine striping disease)	Maize rayado fino virus (MRFV)
20	Maize red leaf and red stripe	Mollicute
	Maize red stripe	Maize red stripe virus (MRSV)
	Maize ring mottle	Maize ring mottle virus (MRMV)
	Maize rio IV	Maize rio cuarto virus (MRCV)
25	Maize rough dwarf (nanismo ruvido)	Maize rough dwarf virus (MRDV) (Cereal tillering disease virus)
	Maize sterile stunt	Maize sterile stunt virus (strains of barley yellow striate virus)
	Maize streak	Maize streak virus (MSV)
30	Maize stripe (maize chlorotic stripe, maize hoja blanca)	Maize stripe virus
	Maize stunting	Maize stunting virus
	Maize tassel abortion	Maize tassel abortion virus (MTAV)
35	Maize vein enation	Maize vein enation virus (MVEV)
	Maize wallaby ear	Maize wallaby ear virus (MWEV)
	Maize white leaf	Maize white leaf virus
	Maize white line mosaic	Maize white line mosaic virus (MWLMV)
40	Millet red leaf	Millet red leaf virus (MRLV)
	Northern cereal mosaic	Northern cereal mosaic virus (NCMV)
	Oat pseudorosette (zakuklivanie)	Oat pseudorosette virus
	Oat sterile dwarf	Oat sterile dwarf virus (OSDV)
45	Rice black-streaked dwarf	Rice black-streaked dwarf virus (RBSDV)
	Rice stripe	Rice stripe virus (RSV)

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	Disease	Pathogen
	Sorghum mosaic	Sorghum mosaic virus (SrMV) (also: sugarcane mosaic virus (SCMV) strains H, I and M)
5	Sugarcane Fiji disease	Sugarcane Fiji disease virus (FDV)
	Sugarcane mosaic	Sugarcane mosaic virus (SCMV) strains A, B, D, E, SC, BC, Sabi and MB (formerly MDMV-B)
10	Wheat spot mosaic	Wheat spot mosaic virus (WSMV)

4. Animal pests

4.1 Insect pathogens:

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Insects such as, for example, beetles, caterpillars, lice or mites may be mentioned by way of example, but not by limitation. Preferred are insects of the genera Coleoptera, Diptera, Hymenoptera, Lepidoptera, Mallophaga, Homoptera, Hemiptera, Orthoptera, Thysanoptera, Dermaptera, Isoptera, Anoplura, Siphonaptera, Trichoptera, and the like. Especially preferred are Coleoptera and Lepidoptera insects such as, for example, the European corn borer (ECB), *Diabrotica barberi* ("northern corn rootworm"), *Diabrotica undecimpunctata* ("southern corn rootworm"), *Diabrotica virgifera* ("Western corn rootworm"), *Agrotis ipsilon* ("black cutworm"), *Crymodes devastator* ("glassy cutworm"), *Feltia ducens* ("dingy cutworm"), *Agrotis gladiaria* ("claybacked cutworm"), *Melanotus* spp., *Aeolus mellillus* ("wireworm"), *Aeolus mancus* ("wheat wireworm"), *Horistonotus uhlerii* ("sand wireworm"), *Sphenophorus maidis* ("maize billbug"), *Sphenophorus zeae* ("timothy billbug"), *Sphenophorus parvulus* ("bluegrass billbug"), *Sphenophorus callosus* ("southern corn billbug"), *Phylloghaga* spp. ("white grubs"), *Anuraphis maidiradicis* ("corn root aphid"), *Delia platura* ("seedcorn maggot"), *Colaspis brunnea* ("grape colaspis"), *Stenolophus lecontei* ("seedcorn beetle") and *Clivinia impressifrons* ("lender seedcorn beetle").

Others which may be mentioned are: the cereal leaf beetle (*Oulema melanopus*), the frit fly (*Oscinella frit*), wireworms (*Agrotis lineatus*) and aphids (such as, for example, the oat grain aphid *Rhopalosiphum padi*, the blackberry aphid *Sitobion avenae*).

4.2 Nematodes:

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Pathogens and the diseases associated with them may be mentioned by way of example, but not by way of limitation, in table 6.

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Table 6: Parasitic nematodes

	Disease	Pathogenic Nematode
	Awl	Dolichodorus spp., D. heterocephalus
10	Bulb and stem nematode disease; Europe	Ditylenchus dipsaci
	Burrowing	Radopholus similis
	Cyst nematode disease	Heterodera avenae, H. zeae, Punctodera chaltoensis
15	Dagger	Xiphinema spp., X. americanum, X. mediterraneum
	False root-knot	Nacobbus dorsalis
	Lance, Columbia	Hoplolaimus columbus
	Lance	Hoplolaimus spp., H. galeatus
20	Lesion	Pratylenchus spp., P. brachyurus, P. crenatus, P. hexincisus, P. neglectus, P. penetrans, P. scribneri, P. thornei, P. zeae
	Needle	Longidorus spp., L. breviannulatus
25	Ring	Criconemella spp., C. ornata
	Root-knot disease	Meloidogyne spp., M. chitwoodi, M. incognita, M. javanica
	Spiral	Helicotylenchus spp.
	Sting	Belonolaimus spp., B. longicaudatus
30	Stubby-root	Paratrichodorus spp., P. christiei, P. minor, Quinisulcius acutus, Trichodorus spp.
	Stunt	Tylenchorhynchus dubius

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Very especially preferred are Globodera rostochiensis and G. pallida (cyst eelworm on potato, tomato and other Solanaceae), Heterodera schachtii (beet eelworm on sugar and fodder beet, oilseed rape, cabbage and the like), Heterodera avenae (cereal cyst nematode on oat and other cereal species), Ditylenchus dipsaci (stem or bulb eelworm, stem eelworm of rye, oats, maize, clover, tobacco, beet), Anguina tritici (ear-cockle nematode, cockle disease of wheat (spelt, rye), Meloidogyne hapla (root-knot nematode of carrot, cucumber, lettuce, tomato, potato, 45 sugar beet, alfalfa).

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Examples of fungal or viral pathogens which are preferred for the individual varieties are the following:

1. Barley:

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fungal, bacterial and viral pathogens: *Puccinia graminis* f.sp. *hordei* (barley stem rust), *Blumeria* (*Erysiphe*) *graminis* f.sp. *hordei* (Barley Powdery Mildew), barley yellow dwarf virus (BYDV),

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Pathogenic insects / nematodes: *Ostrinia nubilalis* (European corn borer); *Agrotis ipsilon* (black cutworm); *Schizaphis graminum* (greenbug); *Blissus leucopterus leucopterus* (chinch bug); *Acrosternum hilare* (green stink bug); *Euschistus servus* (brown stink bug); *Delia platura* (seedcorn maggot); *Mayetiola destructor* (Hessian fly); *Petrobia latens* (brown wheat mite).

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2. Soybean:

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Fungal, bacterial or viral pathogens: *Phytophthora megasperma* fsp. *glycinea*, *Macrophomina phaseolina*, *Rhizoctonia solani*, *Sclerotinia sclerotiorum*, *Fusarium oxysporum*, *Diaporthe phaseolorum* var. *sojae* (*Phomopsis sojae*), *Diaporthe phaseolorum* var. *caulivora*, *Sclerotium rolfsii*, *Cercospora kikuchii*, *Cercospora sojae*, *Peronospora manchurica*, *Colletotrichum dematium* (*Colletotrichum truncatum*), *Corynespora cassiicola*, *Septoria glycines*, *Phyllosticta sojicola*, *Alternaria alternata*, *Pseudomonas syringae* p.v. *glycinea*, *Xanthomonas campestris* p.v. *phaseoli*, *Microsphaera diffusa*, *Fusarium semitectum*, *Phialophora gregata*, Soybean mosaic virus, *Glomerella glycines*, Tobacco Ring spot virus, Tobacco Streak virus, *Phakopsora pachyrhizi*, *Pythium aphanidermatum*, *Pythium ultimum*, *Pythium debaryanum*, Tomato spotted wilt virus, *Heterodera glycines*, *Fusarium solani*.

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Pathogenic insects / nematodes: *Pseudoplusia includens* (soybean looper); *Anticarsia gemmatilis* (velvetbean caterpillar); *Plathypena scabra* (green cloverworm); *Ostrinia nubilalis* (European corn borer); *Agrotis ipsilon* (black cutworm); *Spodoptera exigua* (beet armyworm); *Heliothis virescens* (cotton budworm); *Helicoverpa zea* (cotton bollworm); *Epilachna varivestis* (Mexican bean beetle); *Myzus persicae* (green peach aphid); *Empoasca fabae* (potato leaf hopper); *Acrosternum hilare* (green stink bug); *Melanoplus femurrubrum* (redlegged grasshopper); *Melanoplus differentialis* (differential grasshopper); *Hylemya platura* (seedcorn maggot); *Sericothrips variabilis* (soybean thrips); *Thrips tabaci* (onion thrips); *Te-*

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tranychus turkestan (strawberry spider mite); *Tetranychus urticae* (two-spotted spider mite).

3. Canola:

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Fungal, bacterial or viral pathogens: *Albugo candida*, *Alternaria brassicae*, *Leptosphaeria maculans*, *Rhizoctonia solani*, *Sclerotinia sclerotiorum*, *Mycosphaerella brassicicola*, *Pythium ultimum*, *Peronospora parasitica*, *Fusarium roseum*, *Alternaria alternata*.

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4. Alfalfa:

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Fungal, bacterial or viral pathogens: *Clavibacter michiganense* subsp. *insidiosum*, *Pythium ultimum*, *Pythium irregulare*, *Pythium splendens*, *Pythium debaryanum*, *Pythium aphanidermatum*, *Phytophthora megasperma*, *Peronospora trifoliorum*, *Phoma medicaginis* var. *medicaginis*, *Cercospora medicaginis*, *Pseudopeziza medicaginis*, *Leptotrochila medicaginis*, *Fusarium*, *Xanthomonas campestris* p.v. *alfalfae*, *Aphanomyces euteiches*, *Stemphylium herbarum*, *Stemphylium alfalfae*.

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5. Wheat:

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Fungal, bacterial or viral pathogens: *Pseudomonas syringae* p.v. *atrofaciens*, *Urocystis agropyri*, *Xanthomonas campestris* p.v. *translucens*, *Pseudomonas syringae* p.v. *syringae*, *Alternaria alternata*, *Cladosporium herbarum*, *Fusarium graminearum*, *Fusarium avenaceum*, *Fusarium culmorum*, *Ustilago tritici*, *Ascochyta tritici*, *Cephalosporium gramineum*, *Collotetrachum graminicola*, *Erysiphe graminis* f.sp. *tritici*, *Puccinia graminis* f.sp. *tritici*, *Puccinia recondita* f.sp. *tritici*, *Puccinia striiformis*, *Pyrenophora tritici-repentis*, *Septoria nodorum*, *Septoria tritici*, *Septoria avenae*, *Pseudocercospora herpotrichoides*, *Rhizoctonia solani*, *Rhizoctonia cerealis*, *Gaeumannomyces graminis* var. *tritici*, *Pythium aphanidermatum*, *Pythium arrhenomanes*, *Pythium ultimum*, *Bipolaris sorokiniana*, Barley Yellow Dwarf Virus, Brome Mosaic Virus, Soil-Borne Wheat Mosaic Virus, Wheat Streak Mosaic Virus, Wheat Spindle Streak Virus, American Wheat Striate Virus, *Claviceps purpurea*, *Tilletia tritici*, *Tilletia laevis*, *Ustilago tritici*, *Tilletia indica*, *Rhizoctonia solani*, *Pythium arrhenomanes*, *Pythium graminicola*, *Pythium aphanidermatum*, High Plains Virus, European wheat striate virus, *Puccinia graminis* f.sp. *tritici* (Wheat stem rust), *Blumeria* (*Erysiphe*) *graminis* f.sp. *tritici* (Wheat Powdery Mildew)

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Pathogenic insects / nematodes: *Pseudaletia unipunctata* (army worm); *Spodoptera frugiperda* (fall armyworm); *Elasmopalpus lignosellus* (lesser cornstalk borer); *Agrotis orthogonia* (western cutworm); *Elasmopalpus Zignosellus* (lesser cornstalk borer); *Oulema melanopus* (cereal leaf beetle); *Hypera punctata* (clover leaf weevil); *Diabrotica undecimpunctata howardi* (southern corn rootworm); Russian wheat aphid; *Schizaphis graminum* (greenbug); *Macrosiphum avenae* (English grain aphid); *Melanoplus femurrubrum* (redlegged grasshopper); *Melanoplus differentialis* (differential grasshopper); *Melanoplus sanguinipes* (migratory grasshopper); *Mayetiola destructor* (Hessian fly); *Sitodiplosis mosellana* (wheat midge); *Meromyza americana* (wheat stem maggot); *Hylemya coarctata* (wheat bulb fly); *Frankliniella fusca* (tobacco thrips); *Cephus cinctus* (wheat stem sawfly); *Aceria tulipae* (wheat curl mite).

6. Sunflower:

Fungal, bacterial or viral pathogens: *Plasmophora halstedii*, *Sclerotinia sclerotiorum*, Aster Yellows, *Septoria helianthi*, *Phomopsis helianthi*, *Alternaria helianthi*, *Alternaria zinniae*, *Botrytis cinerea*, *Phoma macdonaldii*, *Macrophomina phaseolina*, *Erysiphe cichoracearum*, *Rhizopus oryzae*, *Rhizopus arrhizus*, *Rhizopus stolonifer*, *Puccinia helianthi*, *Verticillium dahliae*, *Erwinia carotovorum* p.v. *Carotovora*, *Cephalosporium acremonium*, *Phytophthora cryptogea*, *Albugo tragopogonis*.

Pathogenic insects / nematodes: *Suleima helianthana* (sunflower bud moth); *Homoeosoma electellum* (sunflower moth); *zygogramma exclamationis* (sunflower beetle); *Bothyrus gibbosus* (carrot beetle); *Neolasioptera murtfeldtiana* (sunflower seed midge).

35 7. Maize:

Fungal, bacterial or viral pathogens: *Fusarium moniliforme* var. *subglutinans*, *Erwinia stewartii*, *Fusarium moniliforme*, *Gibberella zeae* (*Fusarium graminearum*), *Stenocarpella maydis* (*Diplodia maydis*), *Pythium irregulare*, *Pythium debaryanum*, *Pythium graminicola*, *Pythium splendens*, *Pythium ultimum*, *Pythium aphanidermatum*, *Aspergillus flavus*, *Bipolaris maydis* 0, T (*Cochliobolus heterostrophus*), *Helminthosporium carbonum* I, II & III (*Cochliobolus carbonum*), *Exserohilum turcicum* I, II & III, *Helminthosporium pedicellatum*, *Physoderma maydis*, *Phyllosticta maydis*, *Kabatiella maydis*, *Cercospora sorghi*, *Ustilago maydis*, *Puccinia sorghi*, *Puccinia polysora*, Macro-

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- phomina phaseolina, *Penicillium oxalicum*, *Nigrospora oryzae*, *Cladosporium herbarum*, *Curvularia lunata*, *Curvularia inaequalis*, *Curvularia pallescens*, *Clavibacter michiganense* subsp. *nebraskense*, *Trichoderma viride*, Maize Dwarf Mosaic Virus A & B, Wheat Streak Mosaic Virus, Maize Chlorotic Dwarf Virus, *Claviceps sorghi*, *Pseudomonas avenae*, *Erwinia chrysanthemi* p.v. *Zea*, *Erwinia corotovora*, Corn stunt spiroplasma, *Diplodia macrospora*, *Sclerophthora macrospora*, *Peronosclerospora sorghi*, *Peronosclerospora philippinesis*, *Peronosclerospora maydis*, *Peronosclerospora sacchari*, *Spacelotheca reiliana*, *Phytophthora zeae*, *Cephalosporium maydis*, *Cephalosporium acremonium*, Maize Chlorotic Mottle Virus, High Plains Virus, Maize Mosaic Virus, Maize Rayado Fino Virus, Maize Streak Virus (MSV), Maize Stripe Virus, Maize Rough Dwarf Virus.
- Pathogenic insects / nematodes: *Ostrinia nubilalis* (European corn borer); *Agrotis ipsilon* (black cutworm); *Helicoverpa zea* (corn earworm); *Spodoptera frugiperda* (fall armyworm); *Diatraea grandiosella* (southwestern corn borer); *Elasmopalpus lignosellus* (lesser cornstalk borer); *Diatraea saccharalis* (surgarcane borer); *Diabrotica virgifera* (western corn rootworm); *Diabrotica longicornis barberi* (northern corn rootworm); *Diabrotica undecimpunctata howardi* (southern corn rootworm); *Melanotus* spp. (wireworms); *Cyclocephala borealis* (northern masked chafer; white grub); *Cyclocephala immaculata* (southern masked chafer; white grub); *Popillia japonica* (Japanese beetle); *Chaetocnema pulicaria* (corn flea beetle); *Sphenophorus maidis* (maize billbug); *Rhopalosiphum maidis* (corn leaf aphid); *Anuraphis maidiradicis* (corn root aphid); *Blissus leucopterus leucopterus* (chinch bug); *Melanoplus femurrubrum* (redlegged grasshopper); *Melanoplus sanguinipes* (migratory grasshopper); *Hylemya platura* (seedcorn maggot); *Agromyza parvicornis* (corn blot leafminer); *Anaphothrips obscurus* (grass thrips); *Solenopsis milesta* (thief ant); *Tetranychus urticae* (twospotted spider mite).

8. Sorghum:

- Fungal, bacterial or viral pathogens: *Exserohilum turcicum*, *Colletotrichum graminicola* (*Glomerella graminicola*), *Cercospora sorghi*, *Gloeocercospora sorghi*, *Ascochyta sorghina*, *Pseudomonas syringae* p.v. *syringae*, *Xanthomonas campestris* p.v. *holcicola*, *Pseudomonas andropogonis*, *Puccinia purpurea*, *Macrophomina phaseolina*, *Perconia circinata*, *Fusarium moniliforme*, *Alternaria alternata*, *Bipolaris sorghicola*, *Helminthosporium sorghicola*, *Curvularia lunata*, *Phoma insidiosa*, *Pseudomonas avenae* (*Pseudomonas alboprecipitans*), *Ramulispora*

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- sorghi, *Ramulispora sorghicola*, *Phyllachara sacchari*, *Sporisorium reilianum* (*Sphacelotheca reiliana*), *Sphacelotheca cruenta*, *Sporisorium sorghi*, Sugarcane mosaic H, Maize Dwarf Mosaic Virus A & B, *Claviceps sorghi*, *Rhizoctonia solani*,
 5 *Acremonium strictum*, *Sclerophthora macrospora*, *Peronosclerospora sorghi*, *Peronosclerospora philippinensis*, *Sclerospora graminicola*, *Fusarium graminearum*, *Fusarium oxysporum*, *Pythium arrhenomanes*, *Pythium graminicola*.
- 10 Pathogenic insects / nematodes: *Chilo partellus* (sorghum borer); *Spodoptera frugiperda* (fall armyworm); *Helicoverpa zea* (corn earworm); *Elasmopalpus lignosellus* (lesser cornstalk borer); *Feltia subterranea* (granulate cutworm); *Phyllophaga crinita* (white grub); *Eleodes*, *Conoderus* and *Aeolus* spp.
 15 (wireworm); *Oulema melanopus* (cereal leaf beetle); *Chaetocnema pulicaria* (corn flea beetle); *Sphenophorus maidis* (maize billbug); *Rhopalosiphum maidis* (corn leaf aphid); *Siphaflava* (yellow sugarcane aphid); *Blissus leucopterus leucopterus* (chinch bug); *Contarinia sorghicola* (sorghum midge); *Tetranychus cinnabarinus* (carmine spider mite); *Tetranychus urticae* (two-spotted spider mite).
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9. Cotton:

- 25 Pathogenic insects / nematodes: *Heliothis virescens* (cotton budworm); *Helicoverpa zea* (cotton bollworm); *Spodoptera exigua* (beet armyworm); *Pectinophora gossypiella* (pink bollworm); *Anthonomus grandis grandis* (boll weevil); *Aphis gossypii* (cotton aphid); *Pseudatomoscelis seriatus* (cotton flea-hopper); *Trialeurodes abutilonea* (bandedwinged whitefly); *Lygus lineolaris* (tarnished plant bug); *Melanoplus femurrubrum* (redlegged grasshopper); *Melanoplus differentialis* (differential grasshopper); *Thrips tabaci* (onion thrips); *Frankliniella fusca* (tobacco thrips); *Tetranychus cinnabarinus* (carmine spider mite); *Tetranychus urticae* (twospotted spider mite).
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10. Rice:

- 40 Pathogenic insects / nematodes: *Diatraea saccharalis* (sugarcane borer); *Spodoptera frugiperda* (fall armyworm); *Helicoverpa zea* (corn earworm); *Colaspis brunnea* (grape colaspis); *Lissorhoptrus oryzophilus* (rice water weevil); *Sitophilus oryzae* (rice weevil); *Nephotettix nigropictus* (rice leafhopper); *Blissus leucopterus leucopterus* (chinch bug); *Acrosternum hilare* (green stink bug).
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11. Oilseed rape:

- Pathogenic insects / nematodes: *Brevicoryne brassicae* (cabbage aphid); *Phyllotreta cruciferae* (Flea beetle); *Mamestra conjugrata* (Bertha armyworm); *Plutella xylostella* (Diamond-back moth); *Delia ssp.* (Root maggots).

For the purposes of the invention, "NADPH oxidase" means all those enzymes whose essential characteristic is that they are capable, by means of a single electron transfer, of converting molecular oxygen (O_2) into superoxide (O_2^-). Preferred are those enzymes which are described by the EC class E.C.1.23.45.3. In this context, the NADPH oxidases can consist of one or more polypeptides which may be identical or different.

Preferably, the NADPH oxidase is a flavocytochrome protein and comprises, as prosthetic groups, a cytochrome b and/or an FAD unit. The NADPH oxidase may consist of an $\alpha\beta$ heterodimer, the β subunit being the functional subunit of the flavocytochrome, which may comprise, as glycoprotein, the electron transport components (a hydrophilic, cytosolic, C-terminal domain, comprising NADPH and FAD, and 4 to 6 N-terminal, putative transmembrane α -helices, comprising two histidine-complexed prosthetic heme groups). The α -subunit may comprise a C-terminal, prolin-rich sequence which is capable of binding potential cytosolic, activating factors of the NADPH oxidase. Activation may take place by binding the cytosolic phox proteins (for example p47-phox, p67-phox, p40-phox) and p21rac, a GTP-binding protein.

The skilled worker is familiar with a large number of NADPH oxidases from plant organisms (Torres MA et al. (1998) Plant J 14: 365-370, inter alia). Sequences which may be mentioned by way of example, but not by limitation, are those with the following GenBank Acc. Nos.: AJ251717 (*Hordeum vulgare*), AP003560 (*Oryza sativa* var. *japonica*), AJ320505 (*Nicotiana tabacum*), AB050660 (*Solanum tuberosum*), AF088276 (*Lycopersicon esculentum*), AB008111 (*Arabidopsis thaliana*; Atrboh F), AF055357 (*Arabidopsis thaliana*; RbohD), AJ309006 (*Nicotiana tabacum*; rboh), AP003271 (*Oryza sativa* cv. *japonica*), AF055355 (*Arabidopsis thaliana*; RbohC), AF055353 (*Arabidopsis thaliana*; RbohA). Especially preferred are the NADPH oxidases which comprise a sequence as shown in SEQ ID: 2, 4, 6, 8, 10, 12, 14, 16, 18, 20 or 22.

The sequences from other plants which are homologous to the NADPH oxidase sequences disclosed within the present invention can be found readily for example by database search or by screening genetic libraries using the NADPH oxidase sequences as search se-

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quence or probe. Examples which may be mentioned are sequences with the following GenBank Acc. Nos.: CAC51517.1, AJ251717, T03973, BAB68079.1, AP003560, T02024, CAC87256.1, AJ320505, BAB70750.1, AB050660, AF088276_1, NP_564821.1, NM_105079, T00265
 5 AC007764_16, NP_192862.1, NM_117194, AF147783_1, AAM28891.1, AF506374, CAC84140.1, AJ309006, T51804, NP_199602.1, NM_124165, BAB89740.1, AP003271, AAC39477.1, AF055355, NP_199919.1, NM_124485, AAC39475.1, AF055353, NP_196356.1, NM_120821, NP_194239.1, NM_118641, BAB08369.1, AB015475, AAC39478.1,
 10 AF055356, AC069143_9, NP_173357.1, NM_101781, NP_172383.1, NM_100780, AAB70398.1, AC000106, AAC39476.1, AF055354, BAB70751.1, AB050661, BAB63664.1, AP003275, AAD24966.1, AF109150.

The polypeptide sequence of the NADPH oxidase especially prefer-
 15 ably comprises at least one sequence motif selected from the group of sequence motifs consisting of

- i) AL(K/R)GL(K/R)
- ii) DK(N/D)XDG(R/K)(I/L/V)(T/N)E
- 20 iii) LSASAN
- iv) IMEELDP
- v) K(F/L)NMA(I/L)(I/V)LXPVCRN
- vi) (E/Q)WHPFSIT
- vii) S(A/S)PXDD(Q/Y)(L/I)S(I/V)H(V/I/L)R
- 25 viii) DGPYG(S/A)PAGDY
- ix) L(I/V)GLGIGATP
- x) FYWVTREQGSF
- xi) GVFYCG

30 The peptide sequence very especially preferably comprises at least 2 or 3, very especially preferably at least 4 or 5, most preferably all of the sequence motifs selected from the group of the sequence motifs i), ii), iii), iv), v), vi), vii), viii), ix) x) and xi). (Letters in brackets mean alternative amino acids
 35 which are possible at this position, for example (V/I) means that valine or isoleucine are possible at this position).

NADPH oxidase may also mean any other unit of an NADPH oxidase enzyme complex which is essential for activity of the NADPH oxi-
 40 dase.

"Protein quantity" means the amount of a NADPH oxidase polypeptide in an organism, a tissue, a cell or a cell compartment. "Reduction" of the protein quantity means the quantitative reduction
 45 of the amount of an NADPH oxidase in an organism, a tissue, a cell or a cell compartment - for example by one of the methods described hereinbelow - in comparison with the wild-type of the

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same genus and species to which this method has not been applied, under otherwise identical conditions (such as, for example, culture conditions, age of the plants and the like). In this context, the reduction amounts to at least 10%, preferably at least 5 10% or at least 20%, especially preferably by at least 40% or 60%, very especially preferably by at least 70% or 80%, most preferably by at least 90% or 95%.

"Activity" means the ability of an NADPH oxidase of converting 10 molecular oxygen (O_2) into superoxide (O_2^-). "Reduction" of the activity means the reduction of the total activity of an NADPH oxidase protein in an organism, a tissue, a cell or a cell compartment - for example by one of the methods described hereinbelow - in comparison with the wild type of the same genus and 15 species, to which this method has not been applied, under otherwise identical conditions (such as, for example, culture conditions, age of the plants and the like). In this context, the reduction amounts to at least 10%, preferably at least 10% or at least 20%, especially preferably to at least 40% or 60%, very especially preferably to at least 70% or 80%, most preferably to at 20 least 90% or 95%.

"Function" preferably means the substrate binding capacity of an NADPH oxidase in an organism, a tissue, a cell or a cell compartment. 25 Suitable substrates are low-molecular-weight compounds such as NADPH or FAD, but also the protein interaction partners of an NADPH oxidase.

"Reduction" of the function means, for example, the quantitative 30 reduction of the binding capacity or binding strength of an NADPH oxidase for at least one substrate in an organism, a tissue, a cell or a cell compartment - for example by one of the methods described hereinbelow - in comparison with the wild-type of the same genus and species to which this method has not been applied, 35 under otherwise identical conditions (such as, for example, culture conditions, age of the plants and the like). "Reduction" is also understood as meaning the change in substrate specificity as expressed, for example, by the k_{cat}/K_m value. In this context, the reduction amounts to at least 10%, preferably at least 10% or 40 at least 20%, especially preferably to at least 40% or 60%, very especially preferably to at least 70% or 80%, most preferably to at least 90% or 95%. Binding partners for NADPH oxidase can be identified for example by the yeast-2-hybrid system in the manner with which the skilled worker is familiar.

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- Methods for determining the protein quantity, the activity of NADPH oxidases or the substrate binding capacity are known to the skilled worker. For example, it is possible to measure the NADPH-dependent O_2^- or H_2O_2 production which can be inhibited by DPI (for example via Nitro Blue Tetrazolium [NBT] or cytochrome c reduction). The protein quantity can be determined for example immunologically, using suitable antibodies. Suitable methods are described (Yu L et al. (1999) Blood 94(7):2497-504; Doke N (1983a) Physiol Plant Pathol 23:345-357; Levine A et al. (1994) Cell 79:583-593; Tenhaken R et al. (1995) Proc Nat Acad Sci USA 92: 4158-4163; Sagi M & Fluhr R. (2001) Plant Physiol 126(3):1281-90; Hükelhoven R & Kogel KH (1998) Mol Plant Microbe Interact 11:292-300; and references cited in the above papers).
- 15 "Functional equivalents" of an NADPH oxidase protein preferably means those sequences which are derived from an NADPH oxidase comprising a polypeptide sequence as shown in SEQ ID NO: 2, 4, 6, 8, 10, 12, 14, 16, 18, 20 or 22 or which are homologous with the former and which have the same essential characteristics.
- 20 In this context, the efficiency of the pathogen resistance may deviate both upwards and downwards in comparison with a value obtained when reducing one of the NADPH oxidases comprising a polypeptide sequence as shown in SEQ ID NO: 2, 4, 6, 8, 10, 12, 14, 16, 18, 20 or 22. Preferred functional equivalents are those where the efficiency of the pathogen resistance - measured for example with the aid of the penetration efficiency of a pathogen (development of haustoria) - differs by not more than 50%, preferably 25%, especially preferably 10%, from a comparative value obtained by reducing an NADPH oxidase comprising a polypeptide sequence as described in SEQ ID NO: 2, 4, 6, 8, 10, 12, 14, 16, 18, 20 or 22. Especially preferred are those sequences whose reduction has the result that the efficiency of the pathogen resistance quantitatively exceeds a comparative value obtained by reducing one of the NADPH oxidases comprising a polypeptide sequence as shown in SEQ ID NO: 2, 4, 6, 8, 10, 12, 14, 16, 18, 20 or 22 by more than 50%, preferably 100%, especially preferably 500%, very especially preferably 1000%.
- 40 The comparison is preferably carried out under analogous conditions. "Analogous conditions" means that all framework conditions such as, for example, culture or growth conditions, assay conditions (such as buffer, temperature, substrates, pathogen concentration and the like) between the experiments to be compared are kept identical and that the set-ups differ only by the sequence of the NADPH oxidases to be compared, their organism of origin and, if appropriate, the pathogen. When selecting the
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pathogen for the comparison, the pathogen to be selected for the comparison is that which is most similar to the corresponding other pathogen, taking into consideration the species specificity.

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In particular, "functional equivalents" means natural or artificial mutations of the NADPH oxidases comprising a polypeptide sequence as shown in SEQ ID NO: 2, 4, 6, 8, 10, 12, 14, 16, 18, 20 or 22 and homologous polypeptides from other plants which continue to have essentially identical characteristics. Homologous polypeptides from the above-described preferred plants are preferred. The sequences from other plants (for example *Arabidopsis thaliana*) which are homologous to the NADPH oxidase sequences disclosed within the scope of the present invention can be found readily for example by database search or screening genetic libraries, using the NADPH oxidase sequences as search sequence or probe. Such sequences are detailed above by way of example together with their GenBank Acc No.

20 Mutations comprise substitutions, additions, deletions, inversions or insertions of one or more amino acid residues. Thus, the present invention also comprises for example those polypeptides which are obtained by modification of a polypeptide comprising a polypeptide sequence as shown in SEQ ID NO: 2, 4, 6, 8, 10, 12, 25 14, 16, 18, 20 or 22.

Homology between two nucleic acid sequences is understood as meaning the identity of the two nucleic acid sequences over in each case the entire sequence length which is calculated by comparison with the aid of the program algorithm GAP (Wisconsin Package Version 10.0, University of Wisconsin, Genetics Computer Group (GCG), Madison, USA), setting the following parameters:

Gap weight: 50	Length weight: 3
Average match: 10	Average mismatch: 0

For example a sequence which has at least 80% homology with sequence SEQ ID NO: 1 at the nucleic acid level is understood as meaning a sequence which, upon comparison with the sequence SEQ ID NO: 1 by the above program algorithm with the above parameter set, has at least 80% homology.

Homology between two polypeptides is understood as meaning the identity of the two nucleic acid sequences over in each case the entire sequence length which is calculated by comparison with the aid of the program algorithm GAP (Wisconsin Package Version 10.0,

University of Wisconsin, Genetics Computer Group (GCG), Madison, USA; Altschul et al. (1997) Nucleic Acids Res. 25:3389 et seq.), setting the following parameters:

Length weight: 2

Average mismatch: -2,003

15 Functional equivalents derived from an NADPH oxidase comprising a polypeptide sequence as shown in SEQ ID NO: 2, 4, 6, 8, 10, 12, 14, 16, 18, 20 or 22 by substitution, insertion or deletion have at least 50%, preferably at least 70%, by preference at least 90%, especially preferably at least 95%, very especially prefer-
20 ably at least 98% homology with a polypeptide comprising a polypeptide sequence as shown in SEQ ID NO: 2, 4, 6, 8, 10, 12, 14, 16, 18, 20 or 22 and are distinguished by identical essential characteristics as the former.

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Also, screening cDNA libraries or genomic libraries of other organisms, preferably of the plant species which are mentioned further below as being suitable hosts for the transformation, using the nucleic acid sequences described under SEQ ID NO: 1, 3, 5, 7, 9, 11, 13, 15, 17, 19 or 21 or parts of these as probe, is a method known to the skilled worker for identifying homologs in other species. In this context, the probes derived from the nucleic acid sequences as shown in SEQ ID NO: 1, 3, 5, 7, 9, 11, 13, 15, 17, 19 or 21 have a length of at least 20 bp, preferably at least 50 bp, especially preferably at least 100 bp, very especially preferably at least 200 bp, most preferably at least 400 bp. A DNA strand which is complementary to the sequences de-

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scribed under SEQ ID NO: 1, 3, 5, 7, 9, 11, 13, 15, 17, 19 or 21 may also be employed for screening the libraries.

Functional equivalents comprises DNA sequences which hybridize
5 under standard conditions with the NADPH oxidase nucleic acid sequences described under SEQ ID NO: 1, 3, 5, 7, 9, 11, 13, 15, 17, 19 or 21, the nucleic acid sequence complementary thereto or parts of the above and which, as complete sequences, encode proteins which have the same essential characteristics as a polypep-
10 tide comprising a sequence as shown in SEQ ID NO: 2, 4, 6, 8, 10, 12, 14, 16, 18, 20 or 22.

"Standard hybridization conditions" is to be understood in the broad sense and means stringent or else less stringent
15 hybridization conditions. Such hybridization conditions are described, inter alia, by Sambrook J, Fritsch EF, Maniatis T et al., in Molecular Cloning (A Laboratory Manual), 2nd Edition, Cold Spring Harbor Laboratory Press, 1989, pages 9.31-9.57 or in Current Protocols in Molecular Biology, John Wiley & Sons, N.Y.
20 (1989), 6.3.1-6.3.6.

For example, the conditions during the wash step can be selected from the range of conditions delimited by low-stringency conditions (approximately 2X SSC at 50°C) and high-stringency conditions
25 tions (approximately 0.2X SSC at 50°C, preferably at 65°C) (20X SSC: 0.3M sodium citrate, 3M NaCl, pH 7.0). In addition, the temperature during the wash step can be raised from low-stringency conditions at room temperature, approximately 22°C, to higher-stringency conditions at approximately 65°C. Both of the parameters,
30 salt concentration and temperature, can be varied simultaneously, or else one of the two parameters can be kept constant while only the other is varied. Denaturants, for example formamide or SDS, may also be employed during the hybridization. In the presence of 50% formamide, hybridization is preferably ef-
35 fected at 42°C. Some examples of conditions for hybridization and wash step are shown hereinbelow:

(1) Hybridization conditions can be selected, for example, from the following conditions:

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- a) 4X SSC at 65°C (with - optionally - 100 µg/ml denatured fragmented fish sperm DNA)
- b) 6X SSC at 45°C (with - optionally - 100 µg/ml denatured fragmented fish sperm DNA),
- 45 c) 6X SSC, 0.5% SDS, 50% formamide at 42°C (with - optionally - 100 µg/ml denatured fragmented fish sperm DNA)

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- d) 4X SSC, 50% formamide at 42°C (with - optionally -
100 µg/ml denatured fragmented fish sperm DNA)
e) 2X or 4X SSC at 50°C (low-stringency condition),
f) 30 to 40% formamide, 2X or 4X SSC at 42°C (low-stringency
5 condition).

(2) Wash steps can be selected, for example, from the following conditions:

- 10 a) 0.015 M NaCl/0.0015 M sodium citrate/0.1% SDS at 50°C.
b) 0.1X SSC at 65°C.
c) 0.1X SSC, 0.5% SDS at 68°C.
d) 0.1X SSC, 0.5% SDS, 50% formamide at 42°C.
e) 0.2X SSC, 0.1% SDS at 42°C.
15 f) 2X SSC at 65°C (low-stringency condition).

The reduction of the expression of an NADPH oxidase protein, the NADPH oxidase activity or the NADPH oxidase function can be performed in many ways.

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- "Reduction" or "to reduce" in connection with an NADPH oxidase, an NADPH oxidase activity or NADPH oxidase function is to be interpreted in the broad sense and comprises the partial or essentially complete prevention or blocking (due to a variety of cell-
25 biological mechanisms) of the functionality of an NADPH oxidase in a plant or a part, tissue, organ, cells or seed derived therefrom. A reduction for the purposes of the invention also comprises a quantitative reduction of an NADPH oxidase down to an essentially complete absence of the NADPH oxidase (i.e. lack of
30 detectability of NADPH oxidase activity or NADPH oxidase function, or lack of immunological detectability of the NADPH oxidase protein). In this context, one or more essential units of the NADPH oxidase can be reduced. In this context, the expression of a certain NADPH oxidase or the NADPH oxidase activity or NADPH
35 oxidase function in a cell or an organism is reduced by preferably more than 50%, especially preferably more than 80%, very especially preferably more than 90%.

- A variety of strategies for reducing the expression of an NADPH
40 oxidase protein, the NADPH oxidase activity or NADPH oxidase function are comprised in accordance with the invention. Strategies which may be mentioned by way of example, but not by limitation, are:

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- a) Introducing a double-stranded NADPH oxidase RNA nucleic acid sequence (NAox-dsRNA) or (an) expression cassette(s) ensuring its expression;
- 5 b) Introducing an NADPH oxidase antisense nucleic acid sequence or an expression cassette ensuring its expression. Comprised are those methods in which the antisense nucleic acid sequence is directed against an NADPH oxidase gene (that is to say, genomic DNA sequences) or an NADPH oxidase gene transcript (that is to say, RNA sequences). Also comprised are α -anomeric nucleic acid sequences.
- 10
- c) Introducing an NADPH oxidase antisense nucleic acid sequence in combination with a ribozyme or an expression cassette ensuring its expression
- 15
- d) Introducing NADPH oxidase sense nucleic acid sequences for inducing a cosuppression or an expression cassette ensuring their expression
- 20
- e) Introducing DNA- or protein-binding factors against NADPH oxidase genes, RNAs or proteins or an expression cassette ensuring their expression
- 25 f) Introducing viral nucleic acid sequences and expression constructs bringing about the degradation of NADPH oxidase RNA, or an expression cassette ensuring their expression
- g) Introducing constructs for inducing a homologous recombination at endogenous NADPH oxidase genes, for example for the generation of knock-out mutants.
- 30
- h) Introducing mutations into endogenous NADPH oxidase genes for generating a loss of function (for example generation of stop codons, reading frame shifts and the like)
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Here, each and every one of these methods can bring about a reduction of the NADPH oxidase expression, NADPH oxidase activity or NADPH oxidase function in the sense of the invention. A combined use is also feasible. Further methods are known to the skilled worker and can comprise hindering or preventing the processing of the NADPH oxidase protein, the transport of the NADPH oxidase protein or its mRNA, inhibition of the attachment of ribosomes, inhibition of RNA splicing, induction of an NADPH oxidase RNA degrading enzyme and/or inhibition of the translational elongation or termination.

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The individual methods which are preferred shall be described briefly hereinbelow:

- 5 a) Introducing a double-stranded NADPH oxidase RNA nucleic acid sequence (NAox-dsRNA)

The method of regulating genes by means of double-stranded RNA (double-stranded RNA interference; dsRNAi) has been described many times in animal and plant organisms (for example Matzke MA
10 et al. (2000) Plant Mol Biol 43:401-415; Fire A. et al (1998) Nature 391:806-811; WO 99/32619; WO 99/53050; WO 00/68374; WO 00/44914; WO 00/44895; WO 00/49035; WO 00/63364). The processes and methods described in the abovementioned references are expressly referred to. Efficient gene suppression can also be
15 shown in the case of transient expression or after transient expression, for example as the result of a biolistic transformation (Schweizer P et al. (2000) Plant J 2000 24:895-903). dsRNAi methods are based on the phenomenon that the simultaneous introduction of complementary strand and counterstrand of a gene tran-
20 script brings about a highly-efficient suppression of the expression of the gene in question. The phenotype which results is very similar to a corresponding knock-out mutant (Waterhouse PM et al. (1998) Proc Natl Acad Sci USA 95:13959-64).

25 The dsRNAi method has proved to be particularly efficient and advantageous when reducing the NADPH oxidase expression. As described in WO 99/32619, inter alia, dsRNAi approaches are markedly superior to traditional antisense approaches.

30 A further aspect of the invention therefore relates to double-stranded RNA molecules (dsRNA molecules) which, when introduced into a plant (or a cell, tissue, organ or seed derived therefrom), bring about the reduction of an NADPH oxidase.

35 The double-stranded RNA molecule for reducing the expression of an NADPH oxidase protein comprises

- 40 a) a sense RNA strand comprising at least one ribonucleotide sequence which is essentially identical to at least part of an NADPH oxidase nucleic acid sequence, and
b) an antisense RNA strand which is essentially - preferably completely - complementary to the RNA sense strand of a).

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In a furthermore preferred embodiment, the double-stranded RNA molecule for reducing the expression of an NADPH oxidase protein comprises

- 5 a) a sense RNA strand comprising at least one ribonucleotide sequence which is essentially identical to at least part of the sense RNA transcript of a nucleic acid sequence encoding an NADPH oxidase protein, and
- 10 b) an antisense RNA strand which is essentially - preferably completely - complementary to the RNA sense strand of a).

With regard to the double-stranded RNA molecules, NADPH oxidase nucleic acid sequence preferably means a sequence comprising a sequence as shown in SEQ ID NO: 1, 3, 5, 7, 9, 11, 13, 15, 17, 19 or 21.

"Essentially identical" means that the dsRNA sequence may also have insertions, deletions and individual point mutations in comparison with the NADPH oxidase target sequence or a functional equivalent target sequence while still bringing about an efficient reduction of the expression. Preferably, the homology as defined above between the sense strand of an inhibitory dsRNA and at least part of the sense RNA transcript of a nucleic acid sequence encoding an NADPH oxidase protein or functional equivalent thereof (or between the antisense strand of the complementary strand of a nucleic acid sequence encoding an NADPH oxidase protein or a functional equivalent thereof) amounts to at least 75%, preferably at least 80%, very especially preferably at least 90% most preferably 100%.

The length of the part-segment amounts to at least 10 bases, preferably at least 25 bases, especially preferably at least 50 bases, very especially preferably at least 100 bases, most preferably at least 200 bases or at least 300 bases.

Alternatively, an "essentially identical" dsRNA may also be defined as a nucleic acid sequence which is capable of hybridizing with a part of a storage protein gene transcript (for example in 400 mM NaCl, 40 mM PIPES pH 6.4, 1 mM EDTA at 50°C or 70°C for 12 to 16 h).

"Essentially complementary" means that the antisense RNA strand may also have insertions, deletions and individual point mutations in comparison with the complement of the sense RNA strand. Preferably, the homology between the antisense RNA strand and the complement of the sense RNA strand amounts to at least 80%, pre-

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ferably at least 90%, very especially preferably at least 95%, most preferably 100%.

"Part of the sense RNA transcript" of a nucleic acid sequence encoding an NADPH oxidase protein or a functional equivalent thereof means fragments of an RNA or mRNA transcribed from a nucleic acid sequence encoding an NADPH oxidase protein or a functional equivalent thereof, preferably an NADPH oxidase gene. Here, the fragments preferably have a sequence length of at least 20 bases, preferably at least 50 bases, especially preferably at least 100 bases, very especially preferably at least 200 bases, most preferably at least 500 bases. Also comprised is the complete transcribed RNA or mRNA.

Also comprised is the use of the dsRNA molecules according to the invention in the methods according to the invention for generating a pathogen resistance in plants.

The dsRNA can consist of one or more strands of polymerized ribonucleotides. Furthermore, modifications both of the sugar-phosphate skeleton and of the nucleosides may be present. For example, the phosphodiester bonds of the natural RNA can be modified in such a way that they comprise at least one nitrogen or sulfur heteroatom. Bases can be modified in such a way that the activity of, for example, adenosine deaminase is limited. These and further modifications are described hereinbelow in the methods for stabilizing antisense RNA.

To achieve the same purpose it is, of course, also possible to introduce, into the cell or the organism, a plurality of individual dsRNA molecules, each of which comprises one of the above-defined ribonucleotide sequence segments.

The dsRNA can be produced enzymatically or, fully or in parts, by chemical synthesis.

The double-stranded dsRNA structure can be formed starting from two complementary, separate RNA strands or - preferably - starting from a single, autocomplementary RNA strand.

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In the case of a single, autocomplementary strand, sense and antisense sequence can be linked by a linking sequence (linker) and form for example a hairpin structure. Preferably, the linking sequence may be an intron, which is spliced out after the dsRNA has been synthesized.

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The nucleic acid sequence encoding a dsRNA may comprise further elements such as, for example transcription termination signals or polyadenylation signals.

5 If the two strands of the dsRNA are to be combined in a cell or plant, this may take place in various ways, for example:

- a) transformation of the cell or plant with a vector which comprises both expression cassettes,
- 10 b) cotransformation of the cell or plant with two vectors, where one comprises the expression cassettes with the sense strand and the other comprises the expression cassettes with the antisense strand,
- 15 c) hybridizing two plants, each of which has been transformed with a vector, where one comprises the expression cassettes with the sense strand and the other comprises the expression cassettes with the antisense strand.

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The formation of the RNA duplex can be initiated either outside the cell or within the same. As in WO 99/53050, the dsRNA may also comprise a hairpin structure by linking sense and antisense strand by a linker (for example an intron). The autocomplementary dsRNA structures are preferred since they merely require the expression of a construct and comprise the complementary strands always in an equimolar ratio.

30 The expression cassettes encoding the antisense or sense strand of a dsRNA or the autocomplementary strand of the dsRNA are preferably inserted into a vector and, using the methods described hereinbelow, stably (for example using selection markers) inserted into the genome of a plant in order to ensure durable expression of the dsRNA.

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The dsRNA can be introduced using such an amount that at least one copy per cell is made possible. Larger amounts (for example at least 5, 10, 100, 500 or 1000 copies per cell) may, if appropriate, bring about a more efficient reduction.

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As already described, 100% sequence identity between dsRNA and an NADPH oxidase gene transcript or the gene transcript of a functionally equivalent gene is not necessarily required in order to bring about an efficient reduction of the NADPH oxidase expression. Accordingly, there is the advantage that the method tolerates sequence deviations, as may be present as the result of genetic mutations, polymorphisms or evolutionary divergences. Using

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the dsRNA which has been generated starting from the NADPH oxidase sequence of an organism, it is thus, for example, possible to suppress the NADPH oxidase expression in another organism. The high degree of sequence homology between the NADPH oxidase sequences from rice, maize and barley allows the conclusion that this protein is highly conserved within plants, so that the expression of a dsRNA derived from one of the NADPH oxidase sequences comprising a sequence as shown in SEQ ID NO: 1, 3, 5, 7, 9, 11, 13, 15, 17, 19 or 21 probably also has an advantageous effect in other plant species.

Owing to the high degree of homology between the individual NADPH oxidase proteins and their functional equivalents, it is also possible to suppress the expression of further homologous NADPH oxidase proteins and/or their functional equivalents of the same organism or else the expression of NADPH oxidase proteins in other related species, using a single dsRNA which has been generated starting from a specific NADPH oxidase sequence of an organism. For this purpose, the dsRNA preferably comprises sequence regions of NADPH oxidase gene transcripts which correspond to conserved regions. Said conserved regions can be deduced readily from sequence alignments.

The dsRNA can be synthesized either in vivo or in vitro. To this end, a DNA sequence encoding a dsRNA can be introduced into an expression cassette under the control of at least one genetic control element (such as, for example, promoter, enhancer, silencer, splice donor or acceptor, polyadenylation signal). Suitable advantageous constructions are described hereinbelow. A polyadenylation is not necessary, nor do elements for initiating a translation have to be present.

A dsRNA can be synthesized chemically or enzymatically. To this end, cellular RNA polymerases or bacteriophage RNA polymerases (such as, for example, T3, T7 or SP6 RNA polymerase) can be used. Such methods for the in-vitro expression of RNA are described (WO 97/32016; US 5,593,874; US 5,698,425, US 5,712,135, US 5,789,214, US 5,804,693). A dsRNA which has been synthesized in vitro, either chemically or enzymatically, can be isolated fully or in part from the reaction mixture, for example by extraction, precipitation, electrophoresis, chromatography or combination of these methods, before it is introduced into a cell, tissue or organism. The dsRNA can be introduced directly into the cell or else by applied extracellularly (for example into the interstitial space).

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However, the plant is preferably transformed stably using an expression construct which brings about the expression of the dsRNA. Suitable methods are described hereinbelow.

5 b) Introduction of an NADPH oxidase antisense nucleic acid sequence

Methods for suppressing a particular protein by preventing the accumulation of its mRNA by antisense technology have been described many times, also in plants (Sheehy et al. (1988) Proc Natl Acad Sci USA 85: 8805-8809; US 4,801,340; Mol JN et al. (1990) FEBS Lett 268(2):427-430). The antisense nucleic acid molecule hybridized with, or binds to, the cellular mRNA and/or genomic DNA encoding the NADPH oxidase target protein to be suppressed, which suppresses the transcription and/or translation of the target protein. The hybridization can be brought about in the traditional manner via the formation of a stable duplex or - in the case of genomic DNA - by binding the antisense nucleic acid molecule with the duplex of the genomic DNA by specific interaction in the large groove of the DNA helix.

An antisense nucleic acid sequence suitable for reducing an NADPH oxidase protein can be derived using the nucleic acid sequence which encodes this protein, for example the nucleic acid sequence comprising a sequence as shown in SEQ ID NO: 1, 3, 5, 7, 9, 11, 13, 15, 17, 19 or 21, following the Watson-Crick base pair rules. The antisense nucleic acid sequence can be complementary to all of the transcribed mRNA of said protein, may be limited to the coding region or may consist of one oligonucleotide only, which is complementary to part of the coding or noncoding sequence of the mRNA. Thus, the oligonucleotide may, for example, be complementary to the region which comprises the translation start for said protein. Antisense nucleic acid sequences can have a length of, for example, 5, 10, 15, 20, 25, 30, 35, 40, 45 or 50 nucleotides, but may also be longer and comprise at least 100, 200, 500, 1000, 2000 or 5000 nucleotides. Antisense nucleic acid sequences can be expressed recombinantly or synthesized chemically or enzymatically using methods known to the skilled worker. In the case of chemical synthesis, natural or modified nucleotides may be used. Modified nucleotides can impart an increased biochemical stability to the antisense nucleic acid sequence and may lead to an increased physical stability of the duplex formed of antisense nucleic acid sequence and sense target sequence. Nucleotides which can be used are, for example, phosphorothioate derivatives and acridine-substituted nucleotides such as 5-fluorouracil, 5-bromouracil, 5-chlorouracil, 5-iodouracil, hypoxanthin, xanthin, 4-acetylcytosine, 5-(carboxyhydroxymethyl)uracil,

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5-carboxymethylaminomethyl-2-thiouridine, 5-carboxymethylaminomethyluracil, dihydrouracil, β -D-galactosylqueosine, inosine, N6-isopentenyladenine, 1-methylguanine, 1-methylinosine, 2,2-dimethylguanine, 2-methyladenine, 2-methylguanine, 3-methylcytosine, 5-methylcytosine, N6-adenine, 7-methylguanine, 5-methylaminomethyluracil, 5-methoxyaminomethyl-2-thiouracil, β -D-mannosylqueosine, 5'-methoxycarboxymethyluracil, 5-methoxyuracil, 2-methylthio-N6-isopentenyladenine, uracil-5-oxyacetic acid, pseudouracil, queosine, 2-thiocytosine, 5-methyl-2-thiouracil, 2-thiouracil, 4-thiouracil, 5-methyluracil, uracil-5-oxyacetic acid methyl ester, uracil-5-oxyacetic acid, 5-methyl-2-thiouracil, 3-(3-amino-3-N-2-carboxypropyl)uracil and 2,6-diaminopurine.

In a further preferred embodiment, the expression of an NADPH oxidase protein can be inhibited by nucleotide sequences which are complementary to the regulatory region of an NADPH oxidase gene (for example an NADPH oxidase promoter and/or enhancer) and form triple-helical structures with the DNA double helix therein, so that the transcription of the NADPH oxidase gene is reduced. Suitable methods are described (Helene C (1991) Anticancer Drug Res 6(6):569-84; Helene C et al. (1992) Ann NY Acad Sci 660:27-36; Maher LJ (1992) Bioassays 14(12):807-815).

In a further embodiment, the antisense nucleic acid molecule can be an α -anomeric nucleic acid. Such α -anomeric nucleic acid molecules form specific double-stranded hybrids with complementary RNA in which - in contrast to the conventional β -nucleic acids - the two strands are parallel to one another (Gautier C et al. (1987) Nucleic Acids Res 15:6625-6641). The antisense nucleic acid molecule can furthermore also comprise 2'-O-methylribonucleotides (Inoue et al. (1987) Nucleic Acids Res 15:6131-6148) or chimeric RNA-DNA analogs (Inoue et al. (1987) FEBS Lett 215:327-330).

c) Introduction of an NADPH oxidase antisense nucleic acid sequence in combination with a ribozyme

The above-described antisense strategy can advantageously be coupled with a ribozyme method. Catalytic RNA molecules or ribozymes can be adapted to any target RNA and cleave the phosphodiester backbone at specific positions, whereby the target DNA is functionally deactivated (Tanner NK (1999) FEMS Microbiol Rev 23(3):257-275). The ribozyme itself is not modified thereby, but is capable of cleaving further target RNA molecules in an analogous manner, whereby it gains the properties of an enzyme. The incorporation of ribozyme sequences into antisense RNAs imparts to these antisense-RNAs this enzyme-like, RNA-cleaving property

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and thus increases their efficiency in the inactivation of the target RNA. The preparation and use of suitable ribozyme anti-sense RNA molecules is described for example by Haseloff et al. (1988) Nature 334:585-591.

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In this manner, ribozymes (for example Hammerhead ribozymes; Haselhoff and Gerlach (1988) Nature 334:585-591) can be used for catalytically cleaving the mRNA of an enzyme to be suppressed - for example NADPH oxidase - and for preventing translation. The
10 ribozyme technology can increase the efficiency of an antisense strategy. Methods for the expression of ribozymes for reducing certain proteins are described in (EP 0 291 533, EP 0 321 201, EP 0 360 257). Ribozyme expression in plant cells is also described (Steinecke P et al. (1992) EMBO J 11(4):1525-1530; de
15 Feyter R et al. (1996) Mol Gen Genet. 250(3):329-338). Suitable target sequences and ribozymes can be determined for example as described by "Steinecke P, Ribozymes, Methods in Cell Biology 50, Galbraith et al. eds, Academic Press, Inc. (1995), pp. 449-460", by calculating the secondary structure of ribozyme RNA and target
20 RNA, and by their interaction (Bayley CC et al. (1992) Plant Mol Biol. 18(2):353-361; Lloyd AM and Davis RW et al. (1994) Mol Gen Genet. 242(6):653-657). For example, it is possible to construct derivatives of the Tetrahymena L-19 IVS RNA, which have complementary regions to the mRNA of the NADPH oxidase protein to be
25 suppressed (see also US 4,987,071 and US 5,116,742). Alternatively, such ribozymes can also be identified from a library of diverse ribozymes, using a selection process (Bartel D and Szostak JW (1993) Science 261:1411-1418).

30 d) Introducing an NADPH oxidase sense nucleic acid sequence for inducing a cosuppression

The expression of an NADPH oxidase nucleic acid sequence in sense orientation can lead to a cosuppression of the corresponding
35 homologous, endogenous gene. The expression of sense RNA with homology to an endogenous gene can reduce or switch off the expression of same, similarly to what has been described for anti-sense approaches (Jorgensen et al. (1996) Plant Mol Biol 31(5):957-973; Goring et al. (1991) Proc Natl Acad Sci USA
40 88:1770-1774; Smith et al. (1990) Mol Gen Genet 224:447-481; Napoli et al. (1990) Plant Cell 2:279-289; Van der Krol et al. (1990) Plant Cell 2:291-99). Here, the introduced construct can fully or only partly represent the homologous gene to be reduced. The possibility of translation is not required. The application
45 of this technology to plants is described for example by Napoli et al. (1990) The Plant Cell 2: 279-289 and in US 5,034,323.

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Preferably, cosuppression is carried out using a sequence which is essentially identical to at least a part of the nucleic acid sequence encoding an NADPH oxidase protein or a functional equivalent thereof, for example the nucleic acid sequence comprising a
5 sequence as shown in SEQ ID NO: 1, 3, 5, 7, 9, 11, 13, 15, 17, 19 or 21.

e) Introducing DNA- or protein-binding factors against NADPH oxidase genes, RNAs or proteins

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A reduction of an NADPH oxidase gene expression is also possible using specific DNA-binding factors, for example with factors of the zinc finger transcription factor type. These factors anneal with the genomic sequence of the endogenous target gene, preferably
15 ably in the regulatory regions, and bring about a repression of the endogenous gene. The use of such a method makes it possible to reduce the expression of an endogenous NADPH oxidase gene without it being necessary to recombinantly manipulate the sequence of the latter. Suitable methods for the generation of
20 suitable factors are described (Dreier B et al. (2001) J Biol Chem 276(31):29466-78; Dreier B et al. (2000) J Mol Biol 303(4):489-502; Beerli RR et al. (2000) Proc Natl Acad Sci USA 97 (4):1495-1500; Beerli RR et al. (2000) J Biol Chem 275(42):32617-32627; Segal DJ and Barbas CF 3rd. (2000) Curr
25 Opin Chem Biol 4(1):34-39; Kang JS and Kim JS (2000) J Biol Chem 275(12):8742-8748; Beerli RR et al. (1998) Proc Natl Acad Sci USA 95(25):14628-14633; Kim JS et al. (1997) Proc Natl Acad Sci USA 94(8):3616-3620; Klug A (1999) J Mol Biol 293(2):215-218; Tsai SY et al. (1998) Adv Drug Deliv Rev 30(1-3):23-31; Mapp AK et al.
30 (2000) Proc Natl Acad Sci USA 97(8):3930-3935; Sharrocks AD et al. (1997) Int J Biochem Cell Biol 29(12):1371-1387; Zhang L et al. (2000) J Biol Chem 275(43):33850-33860).

These factors may be selected using any desired portion of an
35 NADPH oxidase gene. Preferably, this segment is located within the promoter region. For gene suppression, however, it may also be located in the region of the coding exons or introns. The corresponding segments are obtainable for the skilled worker by means of database search from the genetic library or - starting
40 from an NADPH oxidase cDNA whose gene is not present in the genetic library, by screening a genomic library for corresponding genomic clones. The methods required for this purpose are known to the skilled worker.

45 Furthermore, it is possible to introduce, into a cell, factors which inhibit the NADPH oxidase target protein itself. The protein-binding factors may be for example aptamers (Famulok M and

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Mayer G (1999) Curr Top Microbiol Immunol 243:123-36) or antibodies or antibody fragments or single-chain antibodies. The way in which these factors are obtained is described and known to the skilled worker. For example, a cytoplasmic scFv antibody has been
5 employed for modulating the activity of the phytochrome A protein in genetically modified tobacco plants (Owen M et al. (1992) Biotechnology (N Y) 10(7):790-794; Franken E et al. (1997) Curr Opin Biotechnol 8(4):411-416; Whitelam (1996) Trend Plant Sci 1:286-272).

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Gene expression can also be suppressed by tailor-made low-molecular-weight synthetic compounds, for example of the polyamide type (Dervan PB and Bürlí RW (1999) Current Opinion in Chemical Biology 3:688-693; Gottesfeld JM et al. (2000) Gene Expr
15 9(1-2):77-91). These oligomers consist of the units 3-(dimethylamino)propylamine, N-methyl-3-hydroxypyrrole, N-methylimidazole and N-methylpyrrole and can be adapted to any portion of double-stranded DNA in such a way that they bind sequence-specifically in the large groove and block the expression of the gene sequences which are located therein. Suitable methods are described (see, inter alia, Bremer RE et al. (2001) Bioorg Med Chem. 9(8):2093-103; Ansari AZ et al. (2001) Chem Biol. 8(6):583-92; Gottesfeld JM et al. (2001) J Mol Biol. 309(3):615-29; Wurtz NR et al. (2001) Org Lett 3(8):1201-3; Wang
25 CC et al. (2001) Bioorg Med Chem 9(3):653-7; Urbach AR and Dervan PB (2001) Proc Natl Acad Sci USA 98(8):4343-8; Chiang SY et al. (2000) J Biol Chem. 275(32):24246-54).

f) Introducing viral nucleic acid sequences and expression
30 constructs which bring about the degradation of NADPH oxidase RNA

NADPH oxidase expression can also be brought about efficiently by inducing the specific NADPH oxidase RNA degradation by the plant
35 with the aid of a viral expression system (amplicon) (Angell, SM et al. (1999) Plant J. 20(3):357-362). These systems - also referred to as "VIGS" (viral induced gene silencing) - introduce nucleic acid sequences with homology to the transcripts to be suppressed into the plant, using viral vectors. Then, transcription is switched off, probably mediated by plant defense mechanisms against viruses. Suitable techniques and methods are described (Ratcliff F et al. (2001) Plant J 25(2):237-45; Fagard M and Vaucheret H (2000) Plant Mol Biol 43(2-3):285-93; Anandalakshmi R et al. (1998) Proc Natl Acad Sci USA 95(22):13079-84;
45 Ruiz MT (1998) Plant Cell 10(6): 937-46).

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g) Introducing constructs for the induction of a homologous recombination on endogenous NADPH oxidase genes, for example for the generation of knock-out mutants.

5 To generate a homologously recombinant organism with reduced NADPH oxidase activity, for example a nucleic acid construct is used which comprises at least a part of the endogenous NADPH oxidase gene which is modified by a deletion, addition or substitution of at least one nucleotide in such a way that the function-
10 ality is reduced or nullified completely. The modification may also affect the regulatory elements (for example the promoter) of the gene, so that the coding sequence remains unaltered, but expression (transcription and/or translation) does not take place and is reduced.

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In the case of conventional homologous recombination, the modified region is flanked at its 5' and 3' end by further nucleic acid sequences which must have a sufficient length for making possible the recombination. The length is, as a rule, in the
20 range of from several hundred bases to several kilobases (Thomas KR and Capecchi MR (1987) Cell 51:503; Strepp et al. (1998) Proc Natl Acad Sci USA 95(8):4368-4373). For the homologous recombination, the host organism - for example a plant - is transformed with the recombination construct using the methods described
25 hereinbelow, and clones which have undergone successful recombination are selected using, for example, an antibiotic or herbicide resistance.

Homologous recombination is a relatively rare event in higher eukaryotes, especially in plants. Random integrations into the host genome predominate. A possibility of removing the randomly integrated sequences and thus of enriching cell clones with a correct homologous recombination consists in using a sequence-specific recombination system as described in US 6,110,736, by which un-
35 specifically integrated sequences can be deleted, which facilitates the selection of events which have integrated successfully via homologous recombination. A multiplicity of sequence-specific recombination systems can be used, examples which may be mentioned being the Cre/lox system of the bacteriophage P1, the FLP/
40 FRT system of yeast, the Gin recombinase of the phage Mu, the Pin recombinase from E. coli and the R/RS system of the plasmid pSR1. Preferred are the bacteriophage P1 Cre/lox and the yeast FLP/FRT system. The FLP/FRT and cre/lox recombinase system has already been employed in plant systems (Odell et al. (1990) Mol Gen Genet
45 223: 369-378)

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h) Introducing mutations into endogenous NADPH oxidase genes for generating a loss of function (for example generation of stop codons, reading frame shifts and the like)

5 Further suitable methods for reducing the NADPH oxidase activity are the introduction of nonsense mutations into endogenous NADPH oxidase genes, for example by means of introducing RNA/DNA oligo-nucleotides into the plant (Zhu et al. (2000) Nat Biotechnol 18(5):555-558), and the generation of knockout mutants with the
10 aid of, for example, T-DNA mutagenesis (Koncz et al. (1992) Plant Mol Biol 20(5):963-976), ENU (N-ethyl-N-nitrosourea) mutagenesis or homologous recombination (Hohn B and Puchta (1999) H Proc Natl Acad Sci USA 96:8321-8323). Point mutations can also be generated by means of DNA-RNA hybrids, which are also known as chimeraplas-
15 ty (Cole-Strauss et al. (1999) Nucl Acids Res 27(5):1323-1330; Kmiec (1999) Gene Therapy American Scientist 87(3):240-247).

The methods of dsRNAi, cosuppression by means of sense RNA and VIGS (virus-induced gene silencing) are also referred to as post-
20 transcriptional gene silencing (PTGS). PTGS methods, including the reduction of the NADPH oxidase function or activity with dominant-negative NADPH oxidase variants are especially advantageous since the requirements to the homology between the endogenous gene to be suppressed and the transgenically expressed sense or
25 dsRNA nucleic acid sequence (or between the endogenous gene and its dominant-negative variant, respectively) are lower than, for example in the case of a traditional antisense approach. Suitable homology criteria are mentioned in the description of the dsRNAi method and applied generally to PTGS methods or dominant-negative
30 approaches. Owing to the high degree of homology between the NADPH oxidase proteins from maize, rice and barley, a high degree of conservation of these protein in plants can be deduced. Thus, using the NADPH oxidase nucleic acid sequences from barley, maize or rice, it is probably also possible efficiently to suppress the
35 expression of homologous NADPH oxidase proteins in other species, without the isolation and structural elucidation of the NADPH oxidase homologs in these species being necessarily required. This substantially reduces the labor involved. Analogously, using dominant-negative variants of an NADPH oxidase protein from rice,
40 maize or barley, it is presumably also possible efficiently to reduce or suppress the function/activity of its homolog in other plant species.

All substances and compounds which directly or indirectly bring
45 about a reduction of the protein quantity, RNA quantity, gene activity or protein activity of an NADPH oxidase protein, shall hereinbelow be grouped together under the term "anti-NADPH oxi-

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dase" compounds. The term "anti-NADPH oxidase" compound explicitly includes the nucleic acid sequences, peptides, proteins or other factors employed in the above-described methods.

5 For the purposes of the invention, "introduction" comprises all those methods which are suitable for introducing an anti-NADPH oxidase compound directly or indirectly into a plant or a cell, compartment, tissue, organ or seed thereof, or generating it therein. Direct and indirect methods are comprised. The introduc-
10 tion can lead to a transient presence of an anti-NADPH-oxidase compound (for example a dsRNA) or else to a stable presence.

In accordance with the different nature of the above-described approaches, the anti-NADPH-oxidase compound can exert its func-
15 tion directly (for example by insertion into an endogenous NADPH oxidase gene). However, the function can also be exerted indirectly after transcription into an RNA (for example in the case of antisense approaches) or after transcription and translation into a protein (for example binding factors). Both directly and
20 indirectly acting anti-NADPH-oxidase compounds are comprised in accordance with the invention.

"Introducing" comprises for example methods such as transfection, transduction or transformation.

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Thus, for example, anti-NADPH-oxidase compounds also comprise recombinant expression constructs which bring about an expression (i.e. transcription and, if appropriate, translation) of, for example, an NADPH oxidase dsRNA or an NADPH oxidase antisense RNA -
30 preferably in a plant or a part, tissue, organ or seed thereof.

In said expression constructs, a nucleic acid molecule whose expression (transcription and, if appropriate, translation) generates an anti-NADPH-oxidase compound, is preferably in functional
35 linkage with at least one genetic control element (for example a promoter) which ensures expression in an organism, preferably in plants. If the expression construct is to be introduced directly into the plant and the anti-NADPH-oxidase compound (for example the NADPH oxidase dsRNA) is to be generated therein *in plantae*,
40 then plant-specific genetic control elements (for example promoters) are preferred. However, the anti-NADPH-oxidase compound can also be generated in other organisms or *in vitro* and then be introduced into the plant (as described in Example 6 and 7).

Here, preferred control elements are all those prokaryotic or eu-
45 karyotic genetic control elements (for example promoters) which

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permit the expression in the organism selected in each case for the production.

Functional linkage is to be understood as meaning, for example, the sequential arrangement of a promoter with the nucleic acid sequence to be expressed (for example an anti-NAox compound) and, if appropriate, further regulatory elements such as, for example, a terminator in such a way that each of the regulatory elements can fulfill its function when the nucleic acid sequence is expressed recombinantly depending on the arrangement of the nucleic acids into sense or antisense RNA. To this end, direct linkage in the chemical sense is not necessarily required. Genetic control sequences such as, for example, enhancer sequences, can also exert their function on the target sequence from positions which are further away, or indeed from other DNA molecules. Preferred arrangements are those in which the nucleic acid sequence to be expressed recombinantly is positioned behind the sequence acting as promoter, so that the two sequences are linked covalently to each other.

Here, the distance between the promoter sequence and the nucleic acid sequence to be expressed recombinantly is less than 200 base pairs, especially preferably less than 100 base pairs, very especially preferably less than 50 base pairs.

Functional linkage, and an expression cassette, can be generated by means of customary recombination and cloning techniques as are described, for example, in Maniatis T, Fritsch EF and Sambrook J (1989) Molecular Cloning: A Laboratory Manual, Cold Spring Harbor Laboratory, Cold Spring Harbor (NY), in Silhavy TJ, Berman ML and Enquist LW (1984) Experiments with Gene Fusions, Cold Spring Harbor Laboratory, Cold Spring Harbor (NY), in Ausubel FM et al. (1987) Current Protocols in Molecular Biology, Greene Publishing Assoc. and Wiley Interscience and in Gelvin et al. (1990) In: Plant Molecular Biology Manual. However, further sequences which, for example, act as a linker with specific cleavage sites for restriction enzymes or as a signal peptide, may also be positioned between the two sequences. The insertion of sequences may also lead to the expression of fusion proteins. Preferably, the expression cassette, consisting of a linkage of promoter and nucleic acid sequence to be expressed, can exist in a vector-integrated form and be inserted into a plant genome, for example by transformation.

However, an expression cassette also denotes those constructions in which a promoter is placed behind an endogenous NADPH oxidase gene - for example by a homologous recombination - and the

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reduction according to the invention, of an NADPH oxidase protein, is brought about by expressing an antisense NADPH oxidase RNA. Analogously, an anti-NADPH-oxidase compound (for example a nucleic acid compound encoding an NADPH oxidase dsRNA or an NADPH oxidase antisense RNA) can be placed behind an endogenous promoter in such a way that the same effect occurs. Both approaches lead to expression cassettes in the sense of the invention.

The term plant-specific promoters is understood as meaning, in principle, any promoter which is capable of governing the expression of genes, in particular foreign genes, in plants or plant parts, plant cells, plant tissues, or plant cultures. Here, expression may be, for example, constitutive, inducible or development-dependent.

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The following are preferred:

a) Constitutive promoters

Preferred vectors are those which make possible a constitutive expression in plants (Benfey et al. (1989) EMBO J 8:2195-2202). "Constitutive" promoter is understood as meaning those promoters which ensure expression in a large number of, preferably all, tissues over a substantial period of plant development, preferably at all stages of plant development. In particular a plant promoter or a promoter derived from a plant virus are preferably used. Particularly preferred is the promoter of the CaMV cauliflower mosaic virus 35S transcript (Franck et al. (1980) Cell 21:285-294; Odell et al. (1985) Nature 313:810-812; Shewmaker et al. (1985) Virology 140:281-288; Gardner et al. (1986) Plant Mol Biol 6:221-228) or the 19S CaMV promoter (US 5,352,605; WO 84/02913; Benfey et al. (1989) EMBO J 8:2195-2202). Another suitable constitutive promoter is the Rubisco small subunit (SSU) promoter (US 4,962,028), the leguminB promoter (GenBank Acc. No. X03677), the Agrobacterium nopaline synthase promoter, the TR dual promoter, the Agrobacterium OCS (octopine synthase) promoter, the ubiquitin promoter (Holtorf S et al. (1995) Plant Mol Biol 29:637-649), the ubiquitin 1 promoter (Christensen et al. (1992) Plant Mol Biol 18:675-689; Bruce et al. (1989) Proc Natl Acad Sci USA 86:9692-9696), the SmaS promoter, the cinnamyl alcohol dehydrogenase promoter (US 5,683,439), the promoters of the ATPase subunits or the promoter of a proline-rich protein from wheat (WO 91/13991), and further promoters of genes whose constitutive expression in plants is known to the skilled worker. Especially preferred as constitutive promoter is the promoter of the nitrilase-1 (nit1)

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gene from *A. thaliana* (GenBank Acc. No.: Y07648.2, Nucleotide 2456-4340, Hillebrand et al. (1996) Gene 170:197-200).

b) Tissue-specific promoters

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Preferred are furthermore promoters with specificity for the anthers, ovaries, flower, leaves, stems, roots and seeds.

- Seed-specific promoters comprise, for example, the phaseolin promoter (US 5,504,200; Bustos MM et al. (1989) Plant Cell 1(9):839-53), the 2S albumin promoter (Joseffson LG et al. (1987) J Biol Chem 262:12196-12201), the legumin promoter (Shirsat A et al. (1989) Mol Gen Genet 215(2): 326-331), the USP (unknown seed protein) promoter (Bäumlein H et al. (1991) Mol Gen Genet 15 225(3):459-467, the napin promoter (US 5,608,152; Stalberg K et al. (1996) L Planta 199:515-519), the sucrose binding protein promoter (WO 00/26388), the legumin B4 promoter (LeB4; Bäumlein H et al. (1991) Mol Gen Genet 225: 121-128; Bäumlein H et al. (1992) Plant J 2(2):233-239; Fiedler U et al. (1995) 20 Biotechnology (NY) 13(10):1090f), the Arabidopsis oleosin promoter (WO 98/45461), the Brassica Bce4 promoter (WO 91/13980). Further suitable seed-specific promoters are those of the genes encoding the high-molecular-weight glutenin (HMWG), gliadin, branching enzyme, ADP glucose pyrophosphatase (AGPase) or starch 25 synthase. Furthermore preferred promoters are those which permit seed-specific expression in monocots such as maize, barley, wheat, rye, rice and the like. The following can be employed advantageously: the promoter of the lpt2 or lpt1 gene (WO 95/15389, WO 95/23230) or the promoters described in 30 WO 99/16890 (promoters of the hordein gene, the glutelin gene, the oryzin gene, the prolamin gene, the gliadin gene, the glutelin gene, the zein gene, the kasirin gene, or the secalin gene).
- 35 Tuber-, storage-root- or root-specific promoters comprise, for example, the class I patatin promoter (B33), the potato cathepsin D inhibitor promoter.

- Leaf-specific promoters comprise the potato cytosolic FBPase 40 promoter (WO 97/05900), the Rubisco (ribulose-1,5-bisphosphate carboxylase) SSU (small subunit) promoter or the ST-LSI promoter from potato (Stockhaus et al. (1989) EMBO J 8:2445-2451). Very especially preferred are epidermis-specific promoters such as, for example, the OXLP gene (oxalate-oxidase-like protein) 45 promoter (Wei et al. (1998) Plant Mol Biol 36:101-112).

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Flower-specific promoters comprise, for example, the phytoene synthase promoter (WO 92/16635) or the promoter of the P-rr gene (WO 98/22593).

- 5 Anther-specific promoters comprise, for example, the 5126 promoter (US 5,689,049, US 5,689,051), the glob-1 promoter and the γ -zein promoter.

c) Chemically inducible promoters

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- The expression cassettes can also comprise a chemically inducible promoter (review article: Gatz et al. (1997) Annu Rev Plant Physiol Plant Mol Biol 48:89-108), by which the expression of the exogenous gene in the plant at a particular point in time can be controlled. Examples which may be mentioned are the PRP1 promoter (Ward et al. (1993) Plant Mol Biol 22:361-366), a salicylic-acid-inducible promoter (WO 95/19443), a benzenesulfonamide-inducible promoter (EP 0 388 186), a tetracyclin-inducible promoter (Gatz et al. (1992) Plant J 2:397-404), an abscisic-acid-inducible promoter (EP 0 335 528) or an ethanol- or cyclohexanone-inducible promoter (WO 93/21334).

d) Stress- or pathogen-inducible promoters

- 25 Further preferred promoters are those which are induced by biotic or abiotic stress such as, for example, the pathogen-inducible promoter of the PRP1 gene (or *gst1* promoter), for example from potato (WO 96/28561; Ward et al. (1993) Plant Mol Biol 22:361-366), the tomato high-temperature-inducible *hsp70* or *hsp80* promoter (US 5,187,267), the potato low-temperature-inducible alpha-amylase promoter (WO 96/12814) or the light-inducible PPDK promoter. Further pathogen-inducible promoters comprise the flax *Fis1* promoter (WO 96/34949), the *Vst1* promoter (Schubert et al. (1997) Plant Mol Biol 34:417-426) and the tobacco EAS4 sesquiterpene cyclase promoter (US 6,100,451).

- Pathogen-inducible promoters furthermore comprise the promoters of genes which are induced as a consequence of infection by pathogens, such as, for example, genes of PR proteins, SAR proteins, β -1,3-glucanase, chitinase and the like (for example Redolfi et al. (1983) Neth J Plant Pathol 89:245-254; Uknes et al. (1992) Plant Cell 4:645-656; Van Loon (1985) Plant Mol Biol 4:111-116; Marineau et al. (1987) Plant Mol Biol 9:335-342; Matton et al. (1987) Molecular Plant-Microbe Interactions 2:325-342; Somssich et al. (1986) Proc Natl Acad Sci USA 83:2427-2430; Somssich et al. (1988) Mol Gen Genetics 2:93-98; Chen et al. (1996) Plant J 10:955-966; Zhang and Sing (1994) Proc

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Natl Acad Sci USA 91:2507-2511; Warner, et al. (1993) Plant J 3:191-201; Siebertz et al. (1989) Plant Cell 1:961-968(1989).

Also comprised are wounding-inducible promoters such as that of
5 the pinII gene (EP-A 0 375 091; Ryan (1990) Ann Rev Phytopath
28:425-449; Duan et al. (1996) Nat Biotech 14:494-498), of the
wun1 and wun2 gene (US 5,428,148), of the win1 and win2 gene
(Stanford et al. (1989) Mol Gen Genet 215:200-208), of the
systemin gene (McGurl et al. (1992) Science 225:1570-1573), of
10 the WIP1 gene (Rohmeier et al. (1993) Plant Mol Biol 22:783-792;
Eckelkamp et al. (1993) FEBS Letters 323:73-76), of the MPI gene
(Corderok et al. (1994) Plant J 6(2):141-150) and the like.

A source of further pathogen-inducible promoters is the PR gene
15 family. A series of elements in these promoters has proved to be
advantageous. Thus, the region -364 to -288 in the promoter of
PR-2d mediates salicylate specificity (Buchel et al. (1996) Plant
Mol Biol 30, 493-504). The sequence 5'-TCATCTTCTT-3' occurs re-
peatedly in the promoter of the barley β -1,3-glucanase and in more
20 than 30 further stress-induced genes. In tobacco, this region
binds a nuclear protein whose abundance is increased by salicy-
late. The PR-1 promoters from tobacco and Arabidopsis (EP-
A 0 332 104, WO 98/03536) are likewise suitable as pathogen-in-
ducible promoters. Preferred, since especially specifically path-
25 ogen-induced, are the acidic PR-5 (aPR5) promoters from barley
(Schweizer et al. (1997) Plant Physiol 114:79-88) and wheat (Reb-
mann et al. (1991) Plant Mol Biol 16:329-331). aPR5 proteins ac-
cumulate in approximately 4 to 6 hours after pathogen attack and
show only very little background expression (WO 99/66057). An ap-
30 proach for achieving an increased pathogen-induced specificity is
the generation of synthetic promoters from combinations of known
pathogen-responsive elements (Rushton et al. (2002) Plant
Cell 14, 749-762; WO 00/01830; WO 99/66057). Further pathogen-in-
ducible promoters from different species are known to the skilled
35 worker (EP-A 1 165 794; EP-A 1 062 356; EP-A 1 041 148; EP-A
1 032 684).

e) Development-dependent promoters

40 Further suitable promoters are, for example, fruit-maturation-
specific promoters such as, for example, the tomato fruit-matura-
tion-specific promoter (WO 94/21794, EP 409 625). Development-de-
pendent promoters comprise partly the tissue-specific promoters
since individual tissues develop by nature in a development-de-
45 pendent fashion.

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Especially preferred are constitutive promoters and also leaf- and/or stem-specific, pathogen-inducible and epidermis-specific promoters, with pathogen-inducible and epidermis-specific promoters being most preferred.

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Furthermore, further promoters may be linked functionally to the nucleic acid sequence to be expressed, which promoters make possible an expression in further plant tissues or in other organisms, such as, for example, *E. coli* bacteria. Suitable plant
10 promoters are, in principle, all of the above-described promoters.

The nucleic acid sequences present in the expression cassettes according to the invention can be linked operably to further
15 genetic control sequences in addition to a promoter. The term "genetic control sequences" is to be understood in the broad sense and refers to all those sequences which have an effect on the generation or the function of the expression cassette according to the invention. For example, genetic control
20 sequences modify the transcription and translation in prokaryotic or eukaryotic organisms. Preferably, the expression cassette according to the invention comprise the promoter with specificity for the embryonal epidermis and/or the flower 5'-upstream of the nucleic acid sequence in question to be expressed recombinantly,
25 and 3'-downstream a terminator sequence as additional genetic control sequence and, if appropriate, further customary regulatory elements, in each case linked functionally to the nucleic acid sequence to be expressed recombinantly.

30 Genetic control sequences also comprise further promoters, promoter elements or minimal promoters, all of which can modify the expression-governing properties. Thus, for example, the tissue-specific expression may additionally depend on certain stress factors, owing to genetic control sequences. Such elements
35 have been described, for example, for water stress, abscisic acid (Lam E and Chua NH (1991) J Biol Chem 266(26): 17131-17135) and heat stress (Schoffl F et al. (1989) Mol Gen Genetics 217(2-3):246-53).

40 In principle, all natural promoters together with their regulatory sequences such as those mentioned above may be used for the method according to the invention. In addition, synthetic promoters may also be used advantageously.

45 Genetic control sequences furthermore also comprise the 5'-untranslated regions, introns or noncoding 3'-region of genes, such as, for example, the actin-1 intron, or the Adh1-S introns

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1, 2 and 6 (general reference: The Maize Handbook, Chapter 116, Freeling and Walbot, Eds., Springer, New York (1994)). It has been demonstrated that they may play a significant role in the regulation of gene expression. Thus, it has been demonstrated
5 that 5'-untranslated sequences can enhance the transient expression of heterologous genes. Examples of translation enhancers which may be mentioned are the tobacco mosaic virus 5' leader sequence (Gallie et al. (1987) Nucl Acids Res 15:8693-8711) and the like. Furthermore, they may promote tissue
10 specificity (Rouster J et al. (1998) Plant J 15:435-440).

The expression cassette may advantageously comprise one or more of what are known as enhancer sequences, linked functionally to the promoter, which make possible an increased recombinant
15 expression of the nucleic acid sequence. Additional advantageous sequences, such as further regulatory elements or terminators, may also be inserted at the 3' end of the nucleic acid sequences to be expressed recombinantly. One or more copies of the nucleic acid sequences to be expressed recombinantly may be present in
20 the gene construct.

Polyadenylation signals which are suitable as control sequences are plant polyadenylation signals, preferably those which essentially correspond to T-DNA polyadenylation signals from
25 *Agrobacterium tumefaciens*, in particular to gene 3 of the T-DNA (octopine synthase) of the Ti plasmid pTiACHS (Gielen et al. (1984) EMBO J 3:835 et seq.) or functional equivalents thereof. Examples of terminator sequences which are especially suitable are the OCS (octopine synthase) terminator and the NOS (nopaline
30 synthase) terminator.

Control sequences are furthermore to be understood as those which make possible homologous recombination or insertion into the genome of a host organism or which permit removal from the
35 genome. Upon homologous recombination, for example the natural promoter of a particular gene can be exchanged to a promoter with specificity for the embryonal epidermis and/or the flower. Methods such as the cre/lox technology permit a tissue-specific, if appropriate inducible, removal of the expression cassette from
40 the genome of the host organism (Sauer B (1998) Methods. 14(4):381-92). Here, certain flanking sequences (lox sequences), which later make possible a removal by means of the cre recombinase, are added to the target gene.

45 An expression cassette and vectors derived therefrom may comprise further functional elements. The term functional element is to be understood in the broad sense and refers to all those elements

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which have an effect on the generation, amplification or function of the expression cassettes, vectors or transgenic organisms according to the invention. The following may be mentioned by way of example, but not by limitation:

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- a) Selection markers which confer a resistance to metabolism inhibitors (such as 2-deoxyglucose-6-phosphate (WO 98/45456)), antibiotics or biocides, preferably herbicides, such as, for example, kanamycin, G 418, bleomycin, hygromycin or phosphinothricin etc. Especially preferred selection markers are those which confer resistance to herbicides. Examples which may be mentioned are: DNA sequences which encode phosphinothricin acetyl transferases (PAT) and which inactivate glutamin synthase inhibitors (bar and pat genes), 5-enolpyruvylshikimate-3-phosphate synthase genes (EPSP synthase genes), which confer resistance to Glyphosate® (N-(phosphonomethyl)glycine), the gox gene, which encodes Glyphosate®-degrading enzymes (glyphosate oxidoreductase), the deh gene (encoding a dehalogenase which inactivates Dalapon), sulfonylurea- and imidazolinone-inactivating acetolactate synthases, and bxn genes, which encode bromoxynil-degrading nitrilase enzymes, the aasa gene, which confers resistance to the antibiotic apectinomycin, the streptomycin phosphotransferase (spt) gene, which allows resistance to streptomycin, the neomycin phosphotransferase (nptII) gene, which confers resistance to kanamycin or geneticin, the hygromycin phosphotransferase (hpt) gene, which mediates resistance to hygromycin, the acetolactate synthase gene (ALS), which confers resistance to sulfonylurea herbicides (for example mutated ALS variants with, for example, the S4 and/or Hra mutation).
- b) Reporter genes which encode readily quantifiable proteins and, via their color or enzyme activity, make possible an assessment of the transformation efficacy, the site of expression or the time of expression. Very especially preferred in this context are reporter proteins (Schenborn E, Groskreutz D. Mol Biotechnol. 1999; 13(1):29-44) such as the green fluorescent protein (GFP) (Sheen et al. (1995) Plant Journal 8(5):777-784; Haseloff et al. (1997) Proc Natl Acad Sci USA 94(6):2122-2127; Reichel et al. (1996) Proc Natl Acad Sci USA 93(12):5888-5893; Tian et al. (1997) Plant Cell Rep 16:267-271; WO 97/41228; Chui WL et al. (1996) Curr Biol 6:325-330; Leffel SM et al. (1997) Biotechniques. 23(5):912-8), chloramphenicol transferase, a luciferase (Ow et al. (1986) Science 234:856-859; Millar et al. (1992) Plant Mol Biol Rep 10:324-414), the aequorin (Prasher et al.
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(1985) Biochem Biophys Res Commun 126(3):1259-1268), β -galactosidase, R-locus gene (encode a protein which regulates the production of anthocyanin pigments (red coloration) in plant tissue and thus makes possible a direct analysis of the promoter activity without addition of extra adjuvants or chromogenic substrates; Dellaporta et al., In: Chromosome Structure and Function: Impact of New Concepts, 18th Stadler Genetics Symposium, 11:263-282, 1988), with β -glucuronidase being very especially preferred (Jefferson et al., EMBO J. 1987, 6, 3901-3907).

- c) Origins of replication, which ensure amplification of the expression cassettes or vectors according to the invention in, for example, *E. coli*. Examples which may be mentioned are ORI (origin of DNA replication) the pBR322 ori or the P15A ori (Sambrook et al.: Molecular Cloning. A Laboratory Manual, 2nd ed. Cold Spring Harbor Laboratory Press, Cold Spring Harbor, NY, 1989).
- d) Elements which are necessary for *Agrobacterium*-mediated plant transformation, such as, for example, the right or left border of the T-DNA or the vir region.

The introduction of an expression cassette according to the invention into an organism or cells, tissues, organs, parts or seeds thereof (preferably into plants or plant cells, tissue, organs, parts or seeds) can be effected advantageously using vectors which comprise the expression cassettes. The expression cassette can be introduced into the vector (for example a plasmid vector) via a suitable restriction cleavage site. The plasmid formed is first introduced into *E. coli*. Correctly transformed *E. coli* are selected, grown, and the recombinant plasmid is obtained by the methods familiar to the skilled worker. Restriction analysis and sequencing may serve to verify the cloning step.

Examples of vectors may be plasmids, cosmids, phages, viruses or else *agrobacteria*. In an advantageous embodiment, the expression cassette is introduced by means of plasmid vectors. Preferred vectors are those which make possible a stable integration of the expression cassette into the host genome.

The generation of a transformed organism (or of a transformed cell or tissue) requires that the DNA, RNA or protein in question is introduced into the corresponding host cell.

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A multiplicity of methods are available for this procedure, which is termed transformation (or transduction or transfection) (Keown et al. (1990) *Methods in Enzymology* 185:527-537). For example, the DNA or RNA can be introduced directly by microinjection or by bombardment with DNA-coated microparticles. Also, the cell can be permeabilized chemically, for example using polyethylene glycol, so that the DNA can enter the cell by diffusion. The DNA can also be introduced by protoplast fusion with other DNA-containing units such as minicells, cells, lysosomes or liposomes. Another suitable method of introducing DNA is electroporation, where the cells are permeabilized reversibly by an electrical pulse. Suitable methods have been described (for example by Bilang et al. (1991) *Gene* 100:247-250; Scheid et al. (1991) *Mol Gen Genet* 228:104-112; Guerche et al. (1987) *Plant Science* 52:111-116; Neuhauser et al. (1987) *Theor Appl Genet* 75:30-36; Klein et al. (1987) *Nature* 327:70-73; Howell et al. (1980) *Science* 208:1265; Horsch et al. (1985) *Science* 227:1229-1231; DeBlock et al. (1989) *Plant Physiology* 91:694-701; *Methods for Plant Molecular Biology* (Weissbach and Weissbach, eds.) Academic Press Inc. (1988); and *Methods in Plant Molecular Biology* (Schuler and Zielinski, eds.) Academic Press Inc. (1989)).

In plants, the above-described methods of transforming and regenerating plants from plant tissues or plant cells are exploited for transient or stable transformation. Suitable methods are especially protoplast transformation by polyethylene-glycol-induced DNA uptake, the biolistic method with the gene gun, what is known as the particle bombardment method, electroporation, incubation of dry embryos in DNA-containing solution, and microinjection.

In addition to these "direct" transformation techniques, transformation can also be effected by bacterial infection by means of *Agrobacterium tumefaciens* or *Agrobacterium rhizogenes*. The *Agrobacterium*-mediated transformation is best suited to dicotyledonous plant cells. The methods are described, for example, by Horsch et al. (1985) *Science* 225: 1229f.

When *agrobacteria* are used, the expression cassette must be integrated into specific plasmids, either into a shuttle or intermediate vector, or into a binary vector. If a Ti or Ri plasmid is to be used for the transformation, at least the right border, but in most cases the right and left border, of the Ti or Ri plasmid T-DNA is linked to the transgenic expression construct to be introduced in the form of a flanking region.

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Binary vectors are preferably used. Binary vectors are capable of replication both in *E. coli* and in *Agrobacterium*. As a rule, they comprise a selection marker gene for the selection of transformed plants (see above) and a linker or polylinker flanked by the
5 right and left T-DNA border sequence. They can be transformed directly into *Agrobacterium* (Holsters et al. (1978) *Mol Gen Genet* 163:181-187). Apart from the T-DNA region, they can additionally comprise elements such as a selection marker gene for the selection of transformed *E. coli* or *agrobacteria* (e.g. the *nptIII*
10 gene). The *Agrobacterium* which acts as host organism in this case should already contain a plasmid with the *vir* region. The latter is required for transferring the T-DNA to the plant cell. An *Agrobacterium* transformed in this way can be used for transforming plant cells. The use of T-DNA for transforming plant
15 cells has been studied and described intensively (EP 120 516; Hoekema, In: *The Binary Plant Vector System*, Offsetdrukkerij Kanters B.V., Alblasserdam, Chapter V; An et al. (1985) *EMBO J* 4:277-287). Various binary vectors are known, some of which are commercially available such as, for example, pBI101.2 or pBIN19
20 (Clontech Laboratories, Inc. USA).

Further promoters which are suitable for expression in plants are described (Rogers et al. (1987) *Meth in Enzymol* 153:253-277; Schardl et al. (1987) *Gene* 61:1-11; Berger et al. (1989) *Proc*
25 *Natl Acad Sci USA* 86:8402-8406).

Direct transformation techniques are suitable for any organism and cell type.

30 The plasmid used need not meet any particular requirements in the case of the injection or electroporation of DNA or RNA into plant cells. Simple plasmids such as those of the pUC series can be used. If complete plants are to be regenerated from the transformed cells, it is advantageous for an additional
35 selectable marker gene to be located on the plasmid.

Stably transformed cells, i.e. those which contain the introduced DNA integrated into the DNA of the host cell, can be selected from untransformed cells when a selectable marker is part of the
40 DNA introduced. Examples of genes which can act as markers are all those which are capable of conferring resistance to a biocide (for example an antibiotic, herbicide or a metabolism inhibitor such as 2-deoxyglucose-6-phosphate WO 98/45456) (see above). Transformed cells which express such marker genes are capable of
45 surviving in the presence of concentrations of a corresponding antibiotic or herbicide which kill an untransformed wild type. Examples are mentioned above and preferably comprise the bar

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gene, which confers resistance to the herbicide phosphinothricin (Rathore KS et al. (1993) Plant Mol Biol 21(5):871-884), the nptII gene, which confers resistance to kanamycin, the hpt gene, which confers resistance to hygromycin, or the EPSP gene, which confers resistance to the herbicide glyphosate. The selection marker permits the selection of transformed cells from untransformed cells (McCormick et al. (1986) Plant Cell Reports 5:81-84). The resulting plants can be bred and hybridized in the customary fashion. Two or more generations should be grown in order to ensure that the genomic integration is stable and hereditary.

The abovementioned methods are described, for example, in Jenes B et al. (1993) Techniques for Gene Transfer, in: Transgenic Plants, Vol. 1, Engineering and Utilization, edited by SD Kung and R Wu, Academic Press, pp. 128-143 and in Potrykus (1991) Annu Rev Plant Physiol Plant Molec Biol 42:205-225. The construct to be expressed is preferably cloned into a vector which is suitable for the transformation of Agrobacterium tumefaciens, for example pBin19 (Bevan et al. (1984) Nucl Acids Res 12:8711f).

As soon as a transformed plant cell has been generated, a complete plant can be obtained using methods known to the skilled worker. For example, callus cultures are used as starting material. The development of shoot and root can be induced from this as yet undifferentiated cell biomass in a known fashion. The shoots obtained can be planted out and bred.

The skilled worker is familiar with such methods of regenerating intact plants from plant cells and plant parts. Methods to do so are described, for example, by Fennell et al. (1992) Plant Cell Rep. 11: 567-570; Stoeger et al (1995) Plant Cell Rep. 14:273-278; Jahne et al. (1994) Theor Appl Genet 89:525-533.

The method according to the invention can advantageously be combined with further methods which bring about a pathogen resistance (for example against insects, fungi, bacteria, nematodes and the like), stress resistance or another improvement of the plant's properties. Examples are mentioned in Dunwell JM, Transgenic approaches to crop improvement, J Exp Bot. 2000;51 Spec No; pages 487-96, inter alia.

With regard to, for example, a nucleic acid sequence, an expression cassette or a vector comprising said nucleic acid sequence or an organism transformed with said nucleic acid sequence, expression cassette or vector, "transgenic" means all those

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constructs which have been generated by recombinant methods in which either

- a) the NADPH oxidase nucleic acid sequence, or
- 5 b) a genetic control sequence which is functionally linked with the NADPH oxidase nucleic acid sequence, for example a promoter, or
- 10 c) (a) and (b)

are not located in their natural genetic environment or have been modified by recombinant methods, an example of a modification being a substitution, addition, deletion, inversion or insertion of
15 one or more nucleotide residues. Natural genetic environment refers to the natural chromosomal locus in the source organism, or to the presence in a genomic library. In the case of a genomic library, the natural genetic environment of the nucleic acid sequence is preferably at least partially retained. The environment
20 flanks the nucleic acid sequence at at least one side and has a sequence length of at least 50 bp, preferably at least 500 bp, especially preferably at least 1000 bp, very especially preferably at least 5000 bp. A naturally occurring expression cassette - for example the naturally occurring combination of the NADPH
25 oxidase promoter and the corresponding NADPH oxidase gene - becomes a transgenic expression cassette when the latter is modified by nonnatural, synthetic ("artificial") methods such as, for example, mutagenization. Suitable methods are described (US 5,565,350; WO 00/15815; see also above).

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Another aspect of the invention relates to transgenic organisms transformed with at least one nucleic acid sequence, expression cassette or vector according to the invention, and to cells, cell cultures, tissues, parts - such as, for example in the case of
35 plant organisms, leaves, roots and the like - or propagation material derived from such organisms. Organism is to be understood in the broad sense and means prokaryotic and eukaryotic organisms, preferably bacteria, yeasts, fungi, animal and plant organisms.

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The following are preferred:

- a) fungi such as *Aspergillus*, *Eremothecium*, *Trichoderma*, *Ashbya*, *Neurospora*, *Fusarium*, *Beauveria* or further fungi described in
45 Indian Chem Eng. Section B. Vol 37, No 1,2 (1995) on page 15,

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table 6. Especially preferred is the filamentous Hemiascomycete *Ashbya gossypii* or *Eremothecium ashbyii*,

- b) yeasts such as *Candida*, *Saccharomyces*, *Hansenula* or *Pichia*,
5 with *Saccharomyces cerevisiae* or *Pichia pastoris* (ATCC Accession No. 201178) being especially preferred,
- c) plants in accordance with the abovementioned definition for
10 "plants",
- d) vertebrates and invertebrates. Especially preferred vertebrates are nonhuman mammals such as dogs, cats, sheep, goats, chickens, mice, rats, cattle or horses. Preferred animal cells comprise CHO, COS, HEK293 cells. Preferred invertebrates comprise insect cells such as *Drosophila* S2 and *Spartan* Sf9 or Sf21 cells,
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- e) prokaryotic organisms such as Gram-positive or Gram-negative bacteria such as *Acetobacter*, *Gluconobacter*, *Corynebacterium*,
20 *Brevibacterium*, *Bacillus*, *Clostridium*, *Cyanobacter*, *Escherichia* (especially *Escherichia coli*), *Serratia*, *Staphylococcus*, *Aerobacter*, *Alcaligenes*, *Penicillium* or *Klebsiella*.

Host or starting organisms which are preferred as transgenic organisms are especially plants in accordance with the abovementioned definition. Included within the scope of the invention are all genera and species of higher and lower plants of the Plant Kingdom. Furthermore included are the mature plants, seeds, shoots and seedlings, and parts, propagation materials and culture derived from them, for example cell cultures. Mature plants means plants at any developmental stage beyond the seedling stage. Seedling means a young immature plant in an early developmental stage. Plants which are especially preferred as host organisms are plants to which the method according to the invention
30 for obtaining a pathogen resistance in accordance with the abovementioned criteria can be applied. Very especially preferred are monocotyledonous plants such as wheat, oats, millet, barley, rye, maize, rice, buckwheat, sorghum, triticale, spelt, linseed, sugar cane, and dicotyledonous crop plants such as oilseed rape,
40 canola, cress, *Arabidopsis*, cabbages, soybeans, alfalfa, pea, bean plants, peanut, potato, tobacco, tomato, egg plant, capsicum, sunflower, tagetes, lettuce, *Calendula*, melon, pumpkin/squash or zucchini.

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The transgenic organisms can be generated with the above-described methods for the transformation or transfection of organisms.

- 5 A further aspect of the invention relates to the use of transgenic organisms according to the invention and of the cells, cell cultures, parts - such as for example in the case of transgenic plant organisms roots, leaves and the like - and transgenic propagation material such as seeds or fruits derived from these organisms for the production of foodstuffs, feedstuffs, pharmaceuticals or fine chemicals.

Furthermore preferred is a method for the recombinant production of pharmaceuticals or fine chemicals in host organisms, where a host organism is transformed with one of the above-described expression cassettes and this expression cassette comprises one or more structural genes which encode the desired fine chemical or catalyze the biosynthesis of the desired fine chemical, the transformed host organism is cultured, and the desired fine chemical is isolated from the culture medium. This method can be applied widely for fine chemicals such as enzymes, vitamins, amino acids, sugars, fatty acids, natural and synthetic flavorings, aroma substances and colorants. Especially preferred is the production of tocopherols and tocotrienols and of carotenoids. Culturing the transformed host organisms, and the isolation from the host organisms or from the culture medium, are carried out with methods known to the skilled worker. The production of pharmaceuticals, such as, for example, antibodies or vaccines, is described by Hood EE, Jilka JM (1999) Curr Opin Biotechnol 10(4):382-6; Ma JK, Vine ND (1999) Curr Top Microbiol Immunol 236:275-92.

Sequences

- 35 1. SEQ ID NO: 1 nucleic acid sequence encoding a
barley (*Hordeum vulgare*) NADPH oxidase.
2. SEQ ID NO: 2 amino acid sequence encoding a
barley (*Hordeum vulgare*) NADPH oxidase.
- 40 3. SEQ ID NO: 3 nucleic acid sequence encoding a
rice (*Oryza sativa* var. *japonica*)
NADPH oxidase

60

4. SEQ ID NO: 4 amino acid sequence encoding a
rice (*Oryza sativa* var. *japonica*)
NADPH oxidase
- 5 5. SEQ ID NO: 5 nucleic acid sequence encoding a
Nicotiana tabacum NADPH oxidase
6. SEQ ID NO: 6 amino acid sequence encoding a
Nicotiana tabacum NADPH oxidase
- 10 7. SEQ ID NO: 7 nucleic acid sequence encoding a
potato (*Solanum tuberosum*)
NADPH oxidase
- 15 8. SEQ ID NO: 8 amino acid sequence encoding a
potato (*Solanum tuberosum*)
NADPH oxidase
- 20 9. SEQ ID NO: 9 nucleic acid sequence encoding a
tomato (*Lycopersicon esculentum*)
NADPH oxidase
- 25 10. SEQ ID NO: 10 amino acid sequence encoding a
tomato (*Lycopersicon esculentum*)
NADPH oxidase
11. SEQ ID NO: 11 nucleic acid sequence encoding a
NADPH oxidase aus *Arabidopsis thaliana* (RbohF)
- 30 12. SEQ ID NO: 12 amino acid sequence encoding a
NADPH oxidase aus NADPH oxidase *Arabidopsis*
thaliana (RbohF)
- 35 13. SEQ ID NO: 13 nucleic acid sequence encoding an
Arabidopsis thaliana (RbohD) NADPH oxidase
14. SEQ ID NO: 14 amino acid sequence encoding an
Arabidopsis thaliana (RbohD) NADPH oxidase
- 40 15. SEQ ID NO: 15 nucleic acid sequence encoding a
Nicotiana tabacum (rboh) NADPH oxidase
16. SEQ ID NO: 16 amino acid sequence encoding a
Nicotiana tabacum (rboh) NADPH oxidase
- 45

61

17. SEQ ID NO: 17 nucleic acid sequence encoding a
rice (*Oryza sativa* var. *japonica*)
NADPH oxidase
- 5 18. SEQ ID NO: 18 amino acid sequence encoding a
rice (*Oryza sativa* var. *japonica*)
NADPH oxidase
- 10 19. SEQ ID NO: 19 nucleic acid sequence encoding an
Arabidopsis thaliana (RbohC) NADPH oxidase
20. SEQ ID NO: 20 amino acid sequence encoding an
Arabidopsis thaliana (RbohC) NADPH oxidase
- 15 21. SEQ ID NO: 21 nucleic acid sequence encoding an
Arabidopsis thaliana (RbohA) NADPH oxidase
22. SEQ ID NO: 22 amino acid sequence encoding an
Arabidopsis thaliana (RbohA) NADPH oxidase
- 20 23. SEQ ID NO: 23 oligonucleotide primer 5' NAOX
5'-GARCAAGGCTCTTTTGATTG-3'
24. SEQ ID NO: 24 oligonucleotide primer 3' NaOX
25 5'-GAAATGCTCCTTATGGAATTC-3'

Figures

Fig. 1: RNA interference with pNAox-dsRNA reduces the penetration
30 efficiency of powdery mildew of barley BghA6 in barley.

The relative penetration efficiency (RPE) was determined in five
individual experiments with inoculation with *Bgh* from barley cv
Pallas. The RPE is calculated as the difference between the pene-
35 tration efficiency of pNAox-dsRNA-transformed cells and the pene-
tration efficiency of control-dsRNA-transformed cells (here: av-
erage penetration efficiency 38.74%). The percent RPE (% RPE) is
calculated from the RPE minus 1, multiplied by 100.

$$40 \text{ RPE} = \frac{[\text{PE in pNAox-dsRNA-transformed cells}]}{[\text{PE in control-dsRNA transformed cells}]}$$

$$\% \text{ RPE} = 100 * (\text{RPE}-1)$$

45 The columns (1) to (5) represent the % RPE (i.e. the deviation of
the penetration efficiency from the average of the penetration
efficiency of the control) when evaluating at least 100 interac-

62

tion sites for in each case one independent experiment. The column (m) represents the average % RPE of the experiments. The error bar indicates the standard error.

- 5 "Control dsRNA" represents the parallel experiments with a control dsRNA. "pNAox" dsRNA represents the experiments with the dsRNA of the barley NADPH oxidase.

In cells which have been bombarded with pNAox-dsRNA, the % RPE
10 was markedly (significance $p=0.0054$) reduced in comparison with cells bombarded with a control dsRNA (TR: human thyroid receptor dsRNA).

Examples

15

General methods:

The chemical synthesis of oligonucleotides can be effected for example in the known manner by the phosphoramidite method (Voet,
20 Voet, 2nd edition, Wiley Press New York, pages 896-897). The cloning steps carried out within the scope of the present invention, such as, for example, restriction cleavages, agarose gel electrophoresis, purification of DNA fragments, transfer of nucleic acids to nitrocellulose and nylon membranes, linking DNA
25 fragments, transformation of *E. coli* cells, bacterial cultures, phage multiplication and sequence analysis of recombinant DNA, are carried out as described by Sambrook et al. (1989) Cold Spring Harbor Laboratory Press; ISBN 0-87969-309-6. Recombinant DNA molecules are sequenced with a laser fluorescence DNA se-
30 quencer from MWG Licor following the method of Sanger (Sanger et al. (1977) Proc Natl Acad Sci USA 74:5463-5467).

Example 1: Plants, pathogens and inoculation

35 The variety Pallas was provided by Lisa Munk, Department of Plant Pathology, Royal Veterinary and Agricultural University, Copenhagen, Denmark. Its production is described (Kølster P et al. (1986) Crop Sci 26: 903-907).

40 Unless otherwise described, the seed, which had been pregerminated for 12 to 36 hours in the dark on damp filter paper, was placed at a rate of 5 kernels along the edge of a square pot (8 X 8 cm) in Fruhstorfer soil, type P, covered with soil and watered regularly with tap water. All plants were grown in controlled-environment cabinets or chambers at 16 to 18°C, 50 to 60%
45 relative atmospheric humidity and a 16-hour-light/8-hour-dark photoperiod at 3000 or 5000 lux (photon flux density 50 or

63

60 $\mu\text{mol s}^{-1}\text{m}^{-2}$) for 5 to 8 days and used in the experiments during the seedling stage. In experiments in which primary leaves were treated, the latter were fully developed.

- 5 Prior to carrying out the transient transfection experiments, the plants were grown in controlled environment cabinets or chambers at 24°C daytime temperature, 20°C nighttime temperature, 50 to 60% relative atmospheric humidity and a 16-hour-light/8-hour-dark photoperiod at 30 000 lux.

10

Powdery mildew of barley *Blumeria graminis* (DC) Speer f.sp. *hordei* Em. Marchal race A6 (Wiberg A (1974) *Hereditas* 77: 89-148) (BghA6) was used for the inoculation of barley plants. The fungus was provided from the Department of Biometry, JLU Gießen. The in-

- 15 oculum was propagated in controlled-environment chambers under identical conditions as described above for the plants by transferring the conidia of infected material to regularly grown 7-day-old barley plants cv. Golden Promise at a density of 100 conidia/mm².

20

The inoculation with BghA6 was carried out using 7-day-old seedlings by shaking off the conidia from infected plants in an inoculation tower at approximately 100 conidia/mm² (unless otherwise specified).

25

Example 2: Cloning of the barley pNAox cDNA sequence

- The cDNA fragments required for the isolation of the HvpNAox cDNA, its cloning, sequencing and generation of probes were ob-
30 tained by RT-PCR using the "One Step RT-PCR Kit" (Life Technologies, Karlsruhe, Germany, or Qiagen, Hilden, Germany). To this end, a total RNA from barley seedlings was used as template. The RNA was isolated from Pallas 3, 5 and 7 days after germination. In addition, RNA was isolated from Pallas and from the back-
35 crossed lines with *mlo5*, *Mlg* or *Mla12* 1, 2 and 5 days after inoculation with *BghA6* on day 7 after germination. The RT-PCR was carried out using primers which are derived from conserved regions of the gp91phox homologs from rice and *Arabidopsis thaliana* (GenBank Acc. No.: X93301 and AB008111):

40

5' NAOX: 5'-GARCAAGGCTCTTTTGATTG-3' (SEQ ID NO: 23) and

3' Naox: 5' GAAATGCTCCTTATGGAATTC 3' (SEQ ID NO: 24)

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In each case 1000 ng of total DNA, 0.4 mM dNTPs, in each case 0.6 mM OPN-1 and OPN-2 primer, 10 µl of RNase inhibitor and 1 µl of enzyme mix in 1x RT buffer (one step RT-PCR Kit, Qiagen, Hilden) were employed for the reaction.

5

The following temperature program is used (PTC-100™ model 96V; MJ Research, Inc., Watertown, Massachusetts):

- | | | |
|----|----|--|
| | 1 | cycle of 30 minutes at 50°C |
| 10 | 1 | cycle of 150 seconds at 94°C |
| | 30 | cycles of 94°C for 45 seconds, 55°C for 1 minute
and 72°C for 2 minutes |
| | 1 | cycle of 72°C for 7 minutes |

- 15 The PCR products were separated by means of 2% w/v agarose gel electrophoresis. This gave a 378 bp RT-PCR product (SEQ ID NO: 1) which encodes a part of the open reading frame of the barley NADPH oxidase. The corresponding cDNA was isolated from an agarose gel and cloned in the pGEM-T vector (Promega, Mannheim, Germany) by means of T-overhang ligation. The cDNAs were sequenced starting from the plasmid DNA using the "Thermo Sequenase Fluorescent Labeled Primer Cycle Sequencing Kit" (Amersham, Freiburg, Germany). The construct was named pGEM-T-pNAox.
- 20

25 Example 3: In-vitro synthesis of the pNAox dsRNA

- The plasmid, which had been employed for the in-vitro RNA transcription, comprises the T7 and SP6 promoters at the respective ends of the inserted nucleic acid sequence, which makes possible the synthesis of sense RNA and antisense RNA. The plasmid can be linearized with suitable restriction enzymes (ApaI for SP6 polymerase and PstI for T7 polymerase) in order to ensure correct transcription of the inserted nucleic acid sequence and to prevent read-through into vectorial sequences. To this end, in each case 10 µg of pGEM-T-pNAox plasmid DNA were cut with ApaI for SP6 polymerase and with and PstI for T7 polymerase. The cut plasmids are extracted in 200 µl of water with the same volume phenol/chloroform/isoamyl alcohol, transferred into a fresh Eppendorf vessel (RNase-free) and centrifuged for 5 minutes at 20 000 g.
- 30
- 40 180 µl of the plasmid solution were treated with 420 µl of ethanol, placed on ice and subsequently precipitated by centrifugation for 30 minutes at 20 000 g and -4°C. The precipitate was taken up in 10 µl of TE buffer. The preparations in question were employed directly in an in-vitro transcription with T7-RNA polymerase and with SP6-RNA polymerase, respectively. RNA polymerases were obtained from Roche Molecular Biology, Mannheim, Germany.
- 45

65

Each transcription mixture contained the following in a volume of 40 μ l:

- 2 μ l linearized plasmid DNA (1 μ g)
- 5 2 μ l NTPs (25 mM) (1.25 mM of each NTP)
- 4 μ l 10x reaction buffer (Roche Molecular Biology),
- 1 μ l RNasin (27 units; Roche Molecular Biology),
- 2 μ l RNA polymerase (40 units)
- 29 μ l DEPC water

10

After 2 hours of incubation at 37°C, in each case some of the reaction mixtures from the transcription of the sense and anti-sense strands were mixed, denatured for 5 minutes at 95°C and thereafter hybridized with one another (annealed) by cooling over 15 30 minutes to a final temperature of 37°C. As an alternative, the mixture of sense and antisense strand can also be cooled for 30 minutes at -20°C after the denaturation. The protein precipitate which formed during denaturation and hybridization was removed by briefly centrifuging at 20 800 g, and the supernatant 20 was used directly for coating tungsten particles (see hereinbelow). For the analysis, in each case 1 μ l of each RNA strand and of the dsRNA were separated on a non-denaturing agarose gel. Successful hybridization is evident by a band shift towards higher molecular weight in comparison with the individual strands.

25

4 μ l of the dsRNA were precipitated with ethanol (by addition of 6 μ l of water, 1 μ l of 3M sodium acetate solution and 25 μ l of ethanol, and centrifugation for at least 5 minutes at 20 000 g and 4°C) and resuspended in 500 μ l of water. The absorption spec- 30 trum between 230 and 300 nm was measured or the absorption at 280 and 260 nm was determined to determine the purity and the concentration of the dsRNA. As a rule, 80 to 100 μ g of dsRNA with an OD₂₆₀/OD₂₈₀ ratio of 1.80 to 1.95 were obtained. If desired, a digestion with DNase I may be carried out, but this has no sub- 35 stantial effect on subsequent results.

The dsRNA of the human thyroid receptor (starting vector pT7beta-Sal (Norman C et al. (1988) Cell 55(6):989-1003), provided by Dr. Baniahmad, Department of Genetics, Gießen, Germany; the sequence 40 of the insert is described under the GenBank Acc. No.: NM_000461) acted as control dsRNA. The plasmid was digested with PvuII to generate the sense RNA and with HindIII to generate the antisense RNA, and the RNA was then transcribed using T7 or SP6 RNA polymerase. The individual process steps for the generation of the 45 control dsRNA are carried out analogously to those described above for the pNAox-dsRNA.

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Example 4: Transient transformation, RNAi and evaluation of the development of the fungal pathogen

Barley cv Pallas leaf segments were transformed with a pNAox
5 dsRNA together with a GFP expression vector. Thereafter the leaves
were inoculated with Bgh and the result was analyzed after 48 h
by means of light and fluorescence microscopy. The penetration
into GFP-expressing cells was assessed by detecting haustoria in
live cells and by assessing the fungal development in precisely
10 those cells. In all five experiments, the bombardment of barley
cv Pallas with pNAox dsRNA resulted in a reduced number of cells
which were successfully penetrated by Bgh in comparison with
cells which had been bombarded with foreign control dsRNA (human
thyroid hormone receptor dsRNA, TR). The resistance-inducing ef-
15 fect of the pNAox dsRNA resulted in an average reduction of the
Bgh penetration efficiency by 35% (Fig. 4).

A method which had already been described for the biolistic
introduction of dsRNA into epidermal cells of barley leaves was
20 employed for the transient transformation (Schweizer P et al.
(1999) Mol Plant Microbe Interact 12:647-54; Schweizer P et al.
(2000) Plant J 2000 24: 895-903). Tungsten particles 1.1 µm in
diameter (particle density 25 mg/ml) were coated with dsRNA
(preparation see above) together with plasmid DNA of the vector
25 pGFP (GFP under the control of the CaMV 35S promoters) as trans-
formation marker. To this end, the following amounts of dsRNA and
reporter plasmid were used for the coating per shot: 1 µg pGFP
and 2 µg dsRNA. Double-stranded RNA was synthesized by annealing
sense and antisense RNA *in vitro* (see above).

30 To prepare microcarriers, 55 mg of tungsten particles (M 17, di-
ameter 1.1 µm; Bio-Rad, Munich) were washed twice with 1 ml of
autoclave-distilled water and once with 1 ml of absolute ethanol,
dried and taken up in 1 ml of 50% strength glycerol (approximate-
35 ly 50 mg/ml stock solution). The solution was diluted with 50%
glycerol to 25 mg/ml, mixed thoroughly prior to use and suspended
in an ultrasonic bath. To coat microcarriers, 1 µg of plasmid,
2 µg of dsRNA (1 µl), 12.5 µl of tungsten particle suspension
(25 mg/ml), 12.5 µl of 1 M Ca(NO₃)₂ solution (pH 10) per shot were
40 combined dropwise with constant mixing, left to stand for 10 min-
utes at RT, centrifuged briefly, and 20 µl of the supernatant
were removed. The remainder with the tungsten particles is resus-
pended (ultrasonic bath) and employed in the experiment.

45 Barley primary leaf segments approximately 4 cm in length were
used. The tissues were placed on 0.5% Phytagar (GibcoBRL™ Life
Technologies™, Karlsruhe) supplemented with 20 µg/ml benzimida-

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zole in Petri dishes (diameter 6.5 cm) and, immediately before the particle bombardment, the edges were covered with a stencil with a rectangular opening of dimensions 2.2 cm × 2.3 cm. One after the other, the dishes were placed on the bottom of the vacuum chamber (Schweizer P et al. (1999) Mol Plant Microbe Interact 12:647-54) over which a nylon mesh (mesh size 0.2 mm, Millipore, Eschborn) had been inserted on a perforated sheet to act as diffuser (5 cm above the bottom, 11 cm underneath the macrocarriers, see hereinbelow) in order to diffuse particle clumps and to slow down the particle stream. The macrocarrier attached at the top of the chamber (plastic sterile filter holder, 13 mm, Gelman Sciences, Swinney, UK) was loaded with 5.8 µl of DNA-coated tungsten particles per shot (microcarriers, see hereinbelow). Using a diaphragm vacuum pump (Vacuubrand, Wertheim), the pressure in the chamber was reduced by 0.9 bar, and the tungsten particles were fired at the surface of the plant tissue at a helium-gas pressure of 9 bar. Immediately thereafter, the chamber was aerated. To label transformed cells, the leaves were bombarded with the plasmid (pGFP; vector on pUC18-basis, CaMV 35S promoter/terminator cassette with inserted GFP gene; Schweizer P et al. (1999) Mol Plant Microbe Interact 12:647-54; provided by Dr. P. Schweizer, Department of Plant Genetics IPK, Gatersleben, Germany). Each time a different plasmid was used for the bombardments, the macrocarrier was cleaned thoroughly with water beforehand. After incubation for four hours after the bombardment with slightly open Petri dishes at RT and with daylight, the leaves were incubated with 100 conidia/mm² of powdery mildew of barley (race A6) and incubated under identical conditions for a further 40 to 48 hours.

Leaf segments were bombarded with the coated particles using a particle inflow gun. For each shot, 312 µg of tungsten particles were applied. 4 hours after the bombardment, the leaves were inoculated with *Blumeria graminis* f.sp. *hordei* mildew (race A6) and, after a further 40 hours, evaluated for symptoms of infection. The result (for example the penetration efficiency, defined as percentage of attacked cells with a mature haustorium and a secondary elongating hypha were analyzed by means of fluorescence and light microscopy. An inoculation with 100 conidia/mm² results in an infection frequency of approximately 50% of the transformed cells. A minimum of 100 interaction sites was evaluated for each individual experiment. Transformed (GFP-expressing) cells were identified under excitation with blue light. Three different categories of transformed cells were distinguished:

68

1. Penetrated cells containing a readily recognizable haustorium. A cell with more than one haustorium was considered as one cell.
- 5 2. Cells which, while attacked by a fungal appressorium, contain no haustorium. A cell which has been attacked more than once by Bgh, but which contains no haustorium, was considered as one cell.
- 10 3. Cells which are not infected by Bgh.

Stomatal cells and guard cells were excluded from the assessment. Surface structures of Bgh were analyzed by means of light microscopy or fluorescence staining of the fungus with 0.1% Calcofluor
 15 (w/v in water) for 30 seconds. The fungal development can be evaluated readily by fluorescence microscopy following staining with Calcofluor. In pNAox-dsRNA-transformed cells, the fungus develops a primary and apressorial germ tube, but no haustorium. The development of a haustorium is a condition for the develop-
 20 ment of a secondary hypha.

The relative penetration efficiency (RPE) is calculated as the difference between the penetration efficiency of transformed cells (transformation with pNAox or control dsRNA) and the pene-
 25 tration efficiency of untransformed cells (here: average penetration efficiency 38.74%). The percent RPE (% RPE) is calculated from the RPE minus 1, multiplied by 100.

$$\text{RPE} = \frac{[\text{PE in pNAox-dsRNA-transformed cells}]}{[\text{PE in control-dsRNA transformed cells}]}$$

30

$$\% \text{ RPE} = 100 * (\text{RPE} - 1)$$

The % RPE value (deviation of the average penetration efficiency
 35 of the control) is used to determine the susceptibility of cells transfected with pNAox-dsRNA (fig. 4).

In the case of the control dsRNA, five different experiments reveal no difference between the transfection with the control
 40 dsRNA and water with regard to the penetration efficiency of Bgh.

To rule out an effect of the dsRNA and the transformation rate or survival rate of the attacked cells, the number of GFP-expressing cells in control experiments and pNAox-dsRNA experiments was
 45 compared. The pNAox-dsRNA had no effect on the total number or the number of the attacked GFP-expressing cells.

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Example 5: NADPH oxidase inhibition with diphenyleneiodonium chloride

The results were supported by further experiments with the NADPH oxidase inhibitor diphenyleneiodonium chloride (DPI; table 1). In general, the experiments were carried out as described by Hückelhoven and Kogel, 1998.

Tab. 1: Effect of DPI on the defense against pathogens in *Pal-*
las^a

Type of interaction	Interactions (% ± standard error)	
	Control ^b	200 µM DPI ^c
Penetration	68.25 ± 9.9	16.25 ± 0.5
Nonpenetration	24.25 ± 6.3	67.5 ± 9.5
HR (Hypersensitive response)	7.5 ± 3.7	16.25 ± 9.3

20

a The DPI treatment was carried out 12 hours after inoculation with the pathogen and the evaluation 36 hours after inoculation.

25

b Controll with 10 mM potassium phosphate buffer, pH 7.8, with DMSO content as in the DPI treatment.

30

c DPI dissolved in 10 mM potassium phosphate buffer, pH 7.8, starting from a 10 mg/ml DPI stock solution in DMSO.

35

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1

SEQUENCE LISTING

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<120> Method for obtaining a pathogen resistance in plants

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  Ile Glu Met His Asn Tyr Leu Thr Ser Val Tyr Glu Glu Gly Asp Ala
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gcc aag ata gga gtg ttc tat tgc gga gct cca gtt ctg gcg cag gaa 289
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            85             90             95

cta agc aac ctt tgc cat gag ttc aat ggc aaa tgc acg aca aaa ttc 337
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            20             25             30

Arg Ser Ala Leu Ile Thr Met Leu Gln Ala Leu Asn His Ala Lys Asn
            35             40             45

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2

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Thr Ile Asn Lys Asp Glu Leu Arg Glu Ile Trp Gln Gln Ile Thr Asp	
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290 295 300	
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Glu Tyr Ala Ala Leu Ile Met Glu Glu Leu Asp Pro Glu Gly Leu Gly	
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Tyr Ile Glu Leu Trp Gln Leu Glu Thr Leu Leu Leu Gln Lys Asp Thr	
325 330 335	
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Tyr Met Asn Tyr Ser Gln Ala Leu Ser Tyr Thr Ser Gln Ala Leu Ser	
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Gln Asn Leu Ala Gly Leu Arg Lys Lys Ser Ser Ile Arg Lys Ile Ser	
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Thr Ser Leu Ser Tyr Tyr Phe Glu Asp Asn Trp Lys Arg Leu Trp Val	
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ctt gca ttg tgg att ggg ata atg gct gga ctg ttc acc tgg aaa ttc	1200
Leu Ala Leu Trp Ile Gly Ile Met Ala Gly Leu Phe Thr Trp Lys Phe	
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Met Gln Tyr Arg Asn Arg Tyr Val Phe Asp Val Met Gly Tyr Cys Val	
405 410 415	
aca aca gca aaa gga gct gct gaa acc cta aag ctg aat atg gca att	1296
Thr Thr Ala Lys Gly Ala Ala Glu Thr Leu Lys Leu Asn Met Ala Ile	
420 425 430	
atc ctc ctg cca gta tgc cgt aac acc att act tgg ttg cga agt aca	1344
Ile Leu Leu Pro Val Cys Arg Asn Thr Ile Thr Trp Leu Arg Ser Thr	
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4

agg gct gca cgg gca cta cct ttt gat gac aac atc aac ttc cac aag	1392
Arg Ala Ala Arg Ala Leu Pro Phe Asp Asp Asn Ile Asn Phe His Lys	
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act att gca gca gca att gtg gtt ggt ata atc ctc cat gca ggg aac	1440
Thr Ile Ala Ala Ala Ile Val Val Gly Ile Ile Leu His Ala Gly Asn	
465 470 475 480	
cac ctt gta tgc gat ttt cca cgg tta ata aaa tca tca gat gag aag	1488
His Leu Val Cys Asp Phe Pro Arg Leu Ile Lys Ser Ser Asp Glu Lys	
485 490 495	
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Tyr Ala Pro Leu Gly Gln Tyr Phe Gly Glu Ile Lys Pro Thr Tyr Phe	
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Thr Leu Val Lys Gly Val Glu Gly Ile Thr Gly Val Ile Met Val Val	
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Cys Met Ile Ile Ala Phe Thr Leu Ala Thr Arg Trp Phe Arg Arg Ser	
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Leu Val Lys Leu Pro Arg Pro Phe Asp Lys Leu Thr Gly Phe Asn Ala	
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Phe Trp Tyr Ser His His Leu Phe Ile Ile Val Tyr Ile Ala Leu Ile	
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Val His Gly Glu Cys Leu Tyr Leu Ile His Val Trp Tyr Arg Arg Thr	
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Thr Trp Met Tyr Leu Ser Val Pro Val Cys Leu Tyr Val Gly Glu Arg	
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Ile Leu Arg Phe Phe Arg Ser Gly Ser Tyr Ser Val Arg Leu Leu Lys	
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Val Ala Ile Tyr Pro Gly Asn Val Leu Thr Leu Gln Met Ser Lys Pro	
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Pro Thr Phe Arg Tyr Lys Ser Gly Gln Tyr Met Phe Val Gln Cys Pro	
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Ala Val Ser Pro Phe Glu Trp His Pro Phe Ser Ile Thr Ser Ala Pro	
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Gly Asp Asp Tyr Leu Ser Ile His Val Arg Gln Leu Gly Asp Trp Thr	
675 680 685	
cga gaa ctc aag aga gta ttt gct gca gct tgt gag ccc cca gcg ggt	2112
Arg Glu Leu Lys Arg Val Phe Ala Ala Ala Cys Glu Pro Pro Ala Gly	
690 695 700	
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Gly Lys Ser Gly Leu Leu Arg Ala Asp Glu Thr Thr Lys Lys Ile Leu	
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5

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ccc aag ctt ctg att gat gga ccg tat ggt tct cct gct cag gat tac 2208
Pro Lys Leu Leu Ile Asp Gly Pro Tyr Gly Ser Pro Ala Gln Asp Tyr
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agc aag tat gat gtt tta tta ctt gtt gga tta gga att ggt gcg aca 2256
Ser Lys Tyr Asp Val Leu Leu Leu Val Gly Leu Gly Ile Gly Ala Thr
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ccc ttt att agc ata tta aaa gat ctt ctg aat aac atc atc aaa atg 2304
Pro Phe Ile Ser Ile Leu Lys Asp Leu Leu Asn Asn Ile Ile Lys Met
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gag gaa gag gag gat gct tct act gat ctt tat cca cca atg ggt cgg 2352
Glu Glu Glu Glu Asp Ala Ser Thr Asp Leu Tyr Pro Pro Met Gly Arg
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aat aag cca cat gtt gat ctg ggc aca ctt atg acg att acc tca aga 2400
Asn Lys Pro His Val Asp Leu Gly Thr Leu Met Thr Ile Thr Ser Arg
      785              790              795      800

cca aag aag atc ttg aag acc aca aat gct tac ttt tac tgg gtg aca 2448
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cgt gag caa ggc tct ttt gat tgg ttc aaa gga gtc atg aat gaa att 2496
Arg Glu Gln Gly Ser Phe Asp Trp Phe Lys Gly Val Met Asn Glu Ile
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gct gac ttg gat caa agg aat atc att gag atg cac aac tac cta aca 2544
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agc gtc tat gag gag ggg gat gcc agg tca gca ctc atc acc atg ctc 2592
Ser Val Tyr Glu Glu Gly Asp Ala Arg Ser Ala Leu Ile Thr Met Leu
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caa gct ctg aac cat gcc aag aat gga gtt gat att gtc tct ggg aca 2640
Gln Ala Leu Asn His Ala Lys Asn Gly Val Asp Ile Val Ser Gly Thr
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aaa gtc cgg aca cat ttt gca cga cca aat tgg aga aag gtc ctt tct 2688
Lys Val Arg Thr His Phe Ala Arg Pro Asn Trp Arg Lys Val Leu Ser
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aaa att tcc tcc aag cat cca tat gcc aaa ata ggt gta ttc tac tgt 2736
Lys Ile Ser Ser Lys His Pro Tyr Ala Lys Ile Gly Val Phe Tyr Cys
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gga gct cca gtc ctg gca caa gaa cta agc aaa ctt tgc cat gaa ttc 2784
Gly Ala Pro Val Leu Ala Gln Glu Leu Ser Lys Leu Cys His Glu Phe
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aac ggg aaa tgc aca acg aag ttc gaa ttc cat aag gag cat ttc tga 2832
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<212> PRT

<213> Oryza sativa

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Gly	Glu	Leu	Thr	Gly	Gly	Pro	Ser	Ser	Ser	Ser	Ser	Arg	Ser	Arg	Ser
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Gly	Leu	Arg	Phe	Ile	Ser	Ser	Asn	Lys	Ala	Asn	Asn	Ala	Trp	Met	Glu
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Val	Gln	Ala	Asn	Phe	Asp	Arg	Leu	Ala	Arg	Asp	Gly	Tyr	Leu	Ser	Arg
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Thr	Ile	Asn	Lys	Asp	Glu	Leu	Arg	Glu	Ile	Trp	Gln	Gln	Ile	Thr	Asp
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Glu	Tyr	Ala	Ala	Leu	Ile	Met	Glu	Glu	Leu	Asp	Pro	Glu	Gly	Leu	Gly
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Tyr	Ile	Glu	Leu	Trp	Gln	Leu	Glu	Thr	Leu	Leu	Leu	Gln	Lys	Asp	Thr
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Tyr	Met	Asn	Tyr	Ser	Gln	Ala	Leu	Ser	Tyr	Thr	Ser	Gln	Ala	Leu	Ser
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Leu	Ala														

Thr	Thr	Ala	Lys	Gly	Ala	Ala	Glu	Thr	Leu	Lys	Leu	Asn	Met	Ala	Ile	
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Ile	Leu	Leu	Pro	Val	Cys	Arg	Asn	Thr	Ile	Thr	Trp	Leu	Arg	Ser	Thr	
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His	Leu	Val	Cys	Asp	Phe	Pro	Arg	Leu	Ile	Lys	Ser	Ser	Asp	Glu	Lys	
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Tyr	Ala	Pro	Leu	Gly	Gln	Tyr	Phe	Gly	Glu	Ile	Lys	Pro	Thr	Tyr	Phe	
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Ala	Val	Ser	Pro	Phe	Glu	Trp	His	Pro	Phe	Ser	Ile	Thr	Ser	Ala	Pro	
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Pro	Lys	Leu	Leu	Ile	Asp	Gly	Pro	Tyr	Gly	Ser	Pro	Ala	Gln	Asp	Tyr	
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Glu	Glu	Glu	Glu	Asp	Ala	Ser	Thr	Asp	Leu	Tyr	Pro	Pro	Met	Gly	Arg	
	770					775					780					
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785					790					795					800	

8

Pro Lys Lys Ile Leu Lys Thr Thr Asn Ala Tyr Phe Tyr Trp Val Thr
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Arg Glu Gln Gly Ser Phe Asp Trp Phe Lys Gly Val Met Asn Glu Ile
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Ala Asp Leu Asp Gln Arg Asn Ile Ile Glu Met His Asn Tyr Leu Thr
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Ser Val Tyr Glu Glu Gly Asp Ala Arg Ser Ala Leu Ile Thr Met Leu
850 855 860

Gln Ala Leu Asn His Ala Lys Asn Gly Val Asp Ile Val Ser Gly Thr
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Lys Val Arg Thr His Phe Ala Arg Pro Asn Trp Arg Lys Val Leu Ser
885 890 895

Lys Ile Ser Ser Lys His Pro Tyr Ala Lys Ile Gly Val Phe Tyr Cys
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<212> DNA

<213> *Nicotiana tabacum*

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<223> coding for NADPH oxidase

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Ser Ser Asp Lys Asp Phe Ser Gly Glu Leu Ser Pro Gly Ala Asp Ser
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Gly Tyr Asn Ser Gly Phe Ala Ser Glu Glu Phe Val Glu Val Thr Leu
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gat ctt cag gat gat gat acc att att cta cgg agc gtt gaa ccg gct 192
Asp Leu Gln Asp Asp Asp Thr Ile Ile Leu Arg Ser Val Glu Pro Ala
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act gtg att aac att gac gct cct gat ctt ccc gcc gga gtc ggt att 240
Thr Val Ile Asn Ile Asp Ala Pro Asp Leu Pro Ala Gly Val Gly Ile
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Ser Gly Val Ser Ile Glu Thr Pro Thr Ser Ala Ser Val Ser Glu Ser
85 90 95

cga tcg ccg acg atc cgc cgg agt tca tct agt aaa ctt cgt cag ttt 336
Arg Ser Pro Thr Ile Arg Arg Ser Ser Ser Ser Lys Leu Arg Gln Phe
100 105 110

9

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Glu	Glu	Tyr	Ala	Ala	Leu	Ile	Met	Glu	Glu	Leu	Asp	Pro	Glu	Arg	Leu	
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Thr	Tyr	Leu	Asn	Tyr	Ser	Gln	Ala	Leu	Ser	Tyr	Thr	Ser	Gln	Ala	Leu	
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10

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Phe	Tyr	Gln	Tyr	Lys	Asn	Lys	Ser	Ala	Phe	Arg	Val	Met	Gly	Tyr	Cys	
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Leu	Ile	Leu	Leu	Pro	Val	Cys	Arg	Asn	Thr	Ile	Thr	Trp	Leu	Arg	Ser	
	450					455						460				
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Thr	Lys	Leu	Ser	His	Phe	Val	Pro	Phe	Asp	Asp	Asn	Ile	Asn	Phe	His	
465					470					475					480	
aag	act	gtc	gct	gca	gcc	att	gtc	act	ggg	atc	ata	ctc	cat	gct	ggg	1488
Lys	Thr	Val	Ala	Ala	Ala	Ile	Val	Thr	Gly	Ile	Ile	Leu	His	Ala	Gly	
				485					490					495		
aac	cat	ctt	gta	tgt	gat	ttc	cca	agg	ctt	ata	cat	gca	gat	gat	caa	1536
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Asp	Tyr	Gln	Ser	Phe	Leu	Ser	Asn	Asp	Phe	Gly	Gln	Ser	Lys	Pro	Gly	
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Tyr	Ile	Asp	Leu	Val	Lys	Gly	Val	Glu	Gly	Val	Thr	Gly	Ile	Ile	Met	
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Arg	Ser	Leu	Ile	Lys	Leu	Pro	Lys	Pro	Phe	Asp	Arg	Leu	Thr	Gly	Phe	
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Asn	Ala	Phe	Trp	Tyr	Ser	His	His	Leu	Leu	Val	Ile	Val	Tyr	Ile	Leu	
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Leu	Ile	Ile	His	Gly	Thr	Phe	Leu	Phe	Leu	Val	His	Lys	Trp	Tyr	Ser	
		595					600					605				
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Lys	Thr	Thr	Trp	Met	Tyr	Leu	Ala	Val	Pro	Val	Leu	Leu	Tyr	Ala	Gly	
			610			615					620					
gaa	aga	act	ctt	aga	ttc	ttc	cgg	tca	ggc	ttg	tac	act	gtc	cgg	ctt	1920
Glu	Arg	Thr	Leu	Arg	Phe	Phe	Arg	Ser	Gly	Leu	Tyr	Thr	Val	Arg	Leu	
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ctg	aaa	gta	gca	ata	tat	cct	gga	aat	gtc	ctc	act	cta	caa	atg	tct	1968
Leu	Lys	Val	Ala	Ile	Tyr	Pro	Gly	Asn	Val	Leu	Thr	Leu	Gln	Met	Ser	
				645					650					655		

11

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Lys	Pro	Pro	Gln	Phe	Arg	Tyr	Lys	Ser	Gly	Gln	Tyr	Met	Phe	Val	Gln	
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Cys	Pro	Ala	Val	Ser	Pro	Phe	Glu	Trp	His	Pro	Phe	Ser	Ile	Thr	Ser	
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Ala	Pro	Gly	Asp	Asp	Tyr	Leu	Ser	Ile	His	Ile	Arg	Gln	Leu	Gly	Asp	
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Trp	Thr	Gln	Glu	Leu	Lys	Arg	Val	Phe	Ser	Glu	Ala	Cys	Glu	Arg	Pro	
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gag	gct	gga	aag	agt	ggc	ctg	ctc	aga	gct	gac	gaa	aac	act	aag	aaa	2208
Glu	Ala	Gly	Lys	Ser	Gly	Leu	Leu	Arg	Ala	Asp	Glu	Asn	Thr	Lys	Lys	
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Ser	Leu	Pro	Lys	Leu	Leu	Ile	Asp	Gly	Pro	Tyr	Gly	Ala	Pro	Ala	Gln	
			740					745					750			
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Ala	Thr	Pro	Phe	Ile	Ser	Ile	Leu	Lys	Asp	Leu	Leu	Val	Asn	Ile	Val	
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Trp	Val	Thr	Arg	Glu	Gln	Gly	Ser	Phe	Asp	Trp	Phe	Lys	Gly	Val	Met	
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Asn	Glu	Val	Ala	Glu	Leu	Asp	Gln	Arg	Gly	Val	Ile	Glu	Met	His	Asn	
	850					855					860					
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Tyr	Leu	Thr	Ser	Val	Tyr	Glu	Glu	Gly	Asp	Ala	Arg	Ser	Ala	Leu	Ile	
	865				870					875					880	
acc	atg	gtc	cag	gca	ctt	aac	cat	gct	aag	aat	ggg	gtt	gat	att	gta	2688
Thr	Met	Val	Gln	Ala	Leu	Asn	His	Ala	Lys	Asn	Gly	Val	Asp	Ile	Val	
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tca	ggc	acc	agg	gtg	agg	aca	cat	ttt	gct	agg	cca	aat	tgg	aag	aaa	2736
Ser	Gly	Thr	Arg	Val	Arg	Thr	His	Phe	Ala	Arg	Pro	Asn	Trp	Lys	Lys	
		900						905					910			
gta	ttt	tcc	aag	acc	tta	acc	aag	cat	gca	aat	gca	aga	ata	ggg	gtt	2784
Val	Phe	Ser	Lys	Thr	Leu	Thr	Lys	His	Ala	Asn	Ala	Arg	Ile	Gly	Val	
		915					920					925				

12

ttc tac tgt ggt gca ccc gta tta gca aaa gaa ctc agc aaa ctc tgc 2832
 Phe Tyr Cys Gly Ala Pro Val Leu Ala Lys Glu Leu Ser Lys Leu Cys
 930 935 940

aaa gag tat aat caa aag ggt gca aca aag ttc gag ttt cac aaa gaa 2880
 Lys Glu Tyr Asn Gln Lys Gly Ala Thr Lys Phe Glu Phe His Lys Glu
 945 950 955 960

cat ttt tag 2889
 His Phe

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<212> PRT

<213> Nicotiana tabacum

<400> 6

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Gly Tyr Asn Ser Gly Phe Ala Ser Glu Glu Phe Val Glu Val Thr Leu
 35 40 45

Asp Leu Gln Asp Asp Asp Thr Ile Ile Leu Arg Ser Val Glu Pro Ala
 50 55 60

Thr Val Ile Asn Ile Asp Ala Pro Asp Leu Pro Ala Gly Val Gly Ile
 65 70 75 80

Ser Gly Val Ser Ile Glu Thr Pro Thr Ser Ala Ser Val Ser Glu Ser
 85 90 95

Arg Ser Pro Thr Ile Arg Arg Ser Ser Ser Ser Lys Leu Arg Gln Phe
 100 105 110

Ser Gln Glu Leu Lys Ala Glu Ala Val Ala Lys Ala Arg Gln Phe Ser
 115 120 125

Gln Glu Leu Lys Ala Glu Leu Arg Arg Phe Ser Trp Ser His Gly His
 130 135 140

Ala Ser Arg Ala Phe Ser Pro Ser Ser Phe Phe Gln Asn Ala Val Val
 145 150 155 160

Gly Thr Gly Asn Gly Val Asp Ser Ala Leu Ala Ala Arg Ala Leu Arg
 165 170 175

Arg Gln Arg Ala Gln Leu Asp Arg Thr Arg Ser Ser Ala His Arg Ala
 180 185 190

Leu Arg Arg Leu Lys Phe Ile Ser Asn Asn Lys Thr Asn Gly Trp Asn
 195 200 205

Glu Val Glu Asn Asn Phe Ser Lys Leu Ala Lys Asp Gly Tyr Leu Tyr
 210 215 220

Arg Ser Asp Phe Ala Gln Cys Ile Gly Met Lys Asp Ser Lys Glu Phe
 225 230 235 240

Ala Leu Glu Leu Phe Asp Ala Leu Ser Arg Arg Arg Arg Leu Lys Val
 245 250 255

Asp Lys Ile Ser Lys Glu Glu Leu Tyr Glu Tyr Trp Ser Gln Ile Thr
 260 265 270

Asp Gln Ser Phe Asp Ser Arg Leu Gln Ile Ser Phe Asp Met Val Asp
 275 280 285

13

Lys	Asn	Glu	Asp	Gly	Arg	Ile	Ala	Glu	Glu	Glu	Val	Lys	Glu	Ile	Ile	290	295	300
Met	Leu	Ser	Ala	Ser	Ala	Asn	Lys	Leu	Ser	Arg	Leu	Lys	Glu	Gln	Ala	305	310	315
Glu	Glu	Tyr	Ala	Ala	Leu	Ile	Met	Glu	Glu	Leu	Asp	Pro	Glu	Arg	Leu	325	330	335
Gly	Tyr	Ile	Glu	Leu	Trp	Gln	Leu	Glu	Thr	Leu	Leu	Leu	Gln	Lys	Asp	340	345	350
Thr	Tyr	Leu	Asn	Tyr	Ser	Gln	Ala	Leu	Ser	Tyr	Thr	Ser	Gln	Ala	Leu	355	360	365
Ser	Gln	Asn	Leu	His	Gly	Leu	Arg	Lys	Lys	Ser	Pro	Ile	Lys	Arg	Met	370	375	380
Ser	Thr	Lys	Leu	Val	Tyr	Ser	Leu	Gln	Glu	Asn	Trp	Lys	Arg	Ile	Trp	385	390	395
Val	Leu	Thr	Leu	Trp	Ile	Leu	Ile	Met	Ile	Gly	Leu	Phe	Leu	Trp	Lys	405	410	415
Phe	Tyr	Gln	Tyr	Lys	Asn	Lys	Ser	Ala	Phe	Arg	Val	Met	Gly	Tyr	Cys	420	425	430
Leu	Val	Thr	Ala	Lys	Gly	Ala	Ala	Glu	Thr	Leu	Lys	Phe	Asn	Met	Ala	435	440	445
Leu	Ile	Leu	Leu	Pro	Val	Cys	Arg	Asn	Thr	Ile	Thr	Trp	Leu	Arg	Ser	450	455	460
Thr	Lys	Leu	Ser	His	Phe	Val	Pro	Phe	Asp	Asp	Asn	Ile	Asn	Phe	His	465	470	475
Lys	Thr	Val	Ala	Ala	Ala	Ile	Val	Thr	Gly	Ile	Ile	Leu	His	Ala	Gly	485	490	495
Asn	His	Leu	Val	Cys	Asp	Phe	Pro	Arg	Leu	Ile	His	Ala	Asp	Asp	Gln	500	505	510
Asp	Tyr	Gln	Ser	Phe	Leu	Ser	Asn	Asp	Phe	Gly	Gln	Ser	Lys	Pro	Gly	515	520	525
Tyr	Ile	Asp	Leu	Val	Lys	Gly	Val	Glu	Gly	Val	Thr	Gly	Ile	Ile	Met	530	535	540
Val	Ile	Leu	Met	Ala	Ile	Ala	Phe	Thr	Leu	Ala	Thr	Arg	Trp	Phe	Arg	545	550	555
Arg	Ser	Leu	Ile	Lys	Leu	Pro	Lys	Pro	Phe	Asp	Arg	Leu	Thr	Gly	Phe	565	570	575
Asn	Ala	Phe	Trp	Tyr	Ser	His	His	Leu	Leu	Val	Ile	Val	Tyr	Ile	Leu	580	585	590
Leu	Ile	Ile	His	Gly	Thr	Phe	Leu	Phe	Leu	Val	His	Lys	Trp	Tyr	Ser	595	600	605
Lys	Thr	Thr	Trp	Met	Tyr	Leu	Ala	Val	Pro	Val	Leu	Leu	Tyr	Ala	Gly	610	615	620
Glu	Arg	Thr	Leu	Arg	Phe	Phe	Arg	Ser	Gly	Leu	Tyr	Thr	Val	Arg	Leu	625	630	635
Leu	Lys	Val	Ala	Ile	Tyr	Pro	Gly	Asn	Val	Leu	Thr	Leu	Gln	Met	Ser	645	650	655
Lys	Pro	Pro	Gln	Phe	Arg	Tyr	Lys	Ser	Gly	Gln	Tyr	Met	Phe	Val	Gln	660	665	670

[illegible]

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15

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Met Arg Gly Leu Pro Gly His	
1 5	
gaa cgc cgg tgg acg tcg gat acg gta tct tcc ggc aag gat tta agt	160
Glu Arg Arg Trp Thr Ser Asp Thr Val Ser Ser Gly Lys Asp Leu Ser	
10 15 20	
ggg gag tca tcg ccg gga act gat tcc ggg aat att tcc ggt ttt gct	208
Gly Glu Ser Ser Pro Gly Thr Asp Ser Gly Asn Ile Ser Gly Phe Ala	
25 30 35	
tcc gag gag ttt gtt gaa gtt ata ctt gat ctt cag gat gat gat acg	256
Ser Glu Glu Phe Val Glu Val Ile Leu Asp Leu Gln Asp Asp Asp Thr	
40 45 50 55	
att att cta cgg agc gtt gaa ccg gct act gta atc aac att gat gct	304
Ile Ile Leu Arg Ser Val Glu Pro Ala Thr Val Ile Asn Ile Asp Ala	
60 65 70	
tct gat cct gct acc gga gtc ggt att ggt gga gta tcg att gaa act	352
Ser Asp Pro Ala Thr Gly Val Gly Ile Gly Gly Val Ser Ile Glu Thr	
75 80 85	
ccg gcg tcg ctg act tcg acg tcg gga act cga tcg ccg acg atg cgt	400
Pro Ala Ser Leu Thr Ser Thr Ser Gly Thr Arg Ser Pro Thr Met Arg	
90 95 100	
cgg agt aca tcg aat aaa tta cgt cag ttt tca cag gag ttg aaa gct	448
Arg Ser Thr Ser Asn Lys Leu Arg Gln Phe Ser Gln Glu Leu Lys Ala	
105 110 115	
gag gct gtc gcg aaa gcg aag cat ttc tcg caa gag ctt aaa gcg gag	496
Glu Ala Val Ala Lys Ala Lys His Phe Ser Gln Glu Leu Lys Ala Glu	
120 125 130 135	
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Leu Arg Arg Phe Ser Trp Ser His Gly His Ala Ser Arg Thr Phe Ser	
140 145 150	
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Pro Ala Ser Phe Phe Gln Asn Ala Val Val Gly Thr Gly Asn Gly Val	
155 160 165	
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Asp Ser Ala Leu Ala Ala Arg Ala Leu Arg Arg Gln Arg Ala Gln Leu	
170 175 180	
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Asp Arg Thr Arg Ser Ser Ala His Lys Ala Leu Arg Gly Leu Lys Phe	
185 190 195	
atc agc aat aac aaa act aac gga tgg aat gaa gtt gaa aac aat ttt	736
Ile Ser Asn Asn Lys Thr Asn Gly Trp Asn Glu Val Glu Asn Asn Phe	
200 205 210 215	
gct aag ctc gct aaa gac ggt tac ctt tat cgc tcc gat ttc gca caa	784
Ala Lys Leu Ala Lys Asp Gly Tyr Leu Tyr Arg Ser Asp Phe Ala Gln	
220 225 230	
tgc atc ggt atg aag gat tca aag gaa ttt gca ttg gaa ttg ttt gat	832
Cys Ile Gly Met Lys Asp Ser Lys Glu Phe Ala Leu Glu Leu Phe Asp	
235 240 245	
gct ttg agt aga aga aga aga ttg aag gtt gat aag att agc aaa gag	880
Ala Leu Ser Arg Arg Arg Arg Leu Lys Val Asp Lys Ile Ser Lys Glu	
250 255 260	

16

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265						270					275					
cgg	ctt	cag	atc	ttc	ttc	gac	atg	gtg	gac	aag	aat	gaa	gat	ggg	cga	976
Arg	Leu	Gln	Ile	Phe	Phe	Asp	Met	Val	Asp	Lys	Asn	Glu	Asp	Gly	Arg	
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Asn	Lys	Leu	Ser	Arg	Leu	Lys	Glu	Gln	Ala	Glu	Glu	Tyr	Ala	Ala	Leu	
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Ile	Met	Glu	Glu	Leu	Asp	Pro	Glu	Arg	Leu	Gly	Tyr	Ile	Glu	Leu	Trp	
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Gln	Leu	Glu	Thr	Leu	Leu	Leu	Gln	Lys	Asp	Thr	Tyr	Leu	Asn	Tyr	Ser	
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Gln	Ala	Leu	Ser	Tyr	Thr	Ser	Gln	Ala	Leu	Ser	Gln	Asn	Leu	Gln	Gly	
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Leu	Arg	Lys	Arg	Ser	Pro	Ile	Arg	Arg	Met	Ser	Thr	Lys	Leu	Val	Tyr	
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Ser	Leu	Gln	Glu	Asn	Trp	Lys	Arg	Ile	Trp	Val	Leu	Val	Leu	Trp	Ile	
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Ala	Ala	Glu	Thr	Leu	Lys	Phe	Asn	Met	Ala	Leu	Ile	Leu	Leu	Pro	Val	
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Cys	Arg	Asn	Thr	Ile	Thr	Phe	Leu	Arg	Ser	Thr	Lys	Leu	Ser	Cys	Phe	
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Phe	Pro	Lys	Leu	Ile	His	Ala	Asn	Asn	Thr	Asn	Tyr	Gln	Lys	Tyr	Leu	
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17

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ttc	ctc	tac	ctt	gtg	cat	aac	tgg	tac	tcc	aaa	acg	aca	tgg	atg	tat	1936
Phe	Leu	Tyr	Leu	Val	His	Asn	Trp	Tyr	Ser	Lys	Thr	Thr	Trp	Met	Tyr	
600					605					610					615	
cta	gca	gtt	cct	gta	ctt	ctc	tac	gca	ggg	gaa	aga	act	ctt	aga	ttc	1984
Leu	Ala	Val	Pro	Val	Leu	Leu	Tyr	Ala	Gly	Glu	Arg	Thr	Leu	Arg	Phe	
			620						625					630		
ttc	cga	tca	ggc	tta	tat	aca	gtc	cgg	ctt	cta	aaa	gta	gca	ata	tat	2032
Phe	Arg	Ser	Gly	Leu	Tyr	Thr	Val	Arg	Leu	Leu	Lys	Val	Ala	Ile	Tyr	
			635					640					645			
cct	gga	aat	gtc	ctt	act	ctg	caa	atg	tct	aag	cct	ccg	caa	ttt	cga	2080
Pro	Gly	Asn	Val	Leu	Thr	Leu	Gln	Met	Ser	Lys	Pro	Pro	Gln	Phe	Arg	
		650					655					660				
tac	aag	agt	gga	caa	tat	atg	ttt	gtc	cag	tgt	cca	gct	gtt	tct	cca	2128
Tyr	Lys	Ser	Gly	Gln	Tyr	Met	Phe	Val	Gln	Cys	Pro	Ala	Val	Ser	Pro	
	665					670					675					
ttc	gag	tgg	cat	cca	ttt	tcc	att	act	tca	gct	cct	ggg	gat	gac	tac	2176
Phe	Glu	Trp	His	Pro	Phe	Ser	Ile	Thr	Ser	Ala	Pro	Gly	Asp	Asp	Tyr	
680					685					690					695	
ttg	agc	att	cat	atc	cga	caa	ctt	ggt	gac	tgg	act	caa	gaa	ctc	aag	2224
Leu	Ser	Ile	His	Ile	Arg	Gln	Leu	Gly	Asp	Trp	Thr	Gln	Glu	Leu	Lys	
				700					705					710		
cgg	gtg	ttt	tcc	gag	gct	tgc	gag	cag	cca	gag	gct	gga	aag	agt	ggc	2272
Arg	Val	Phe	Ser	Glu	Ala	Cys	Glu	Gln	Pro	Glu	Ala	Gly	Lys	Ser	Gly	
			715					720					725			
ctg	ctc	aga	gct	gac	gaa	aac	acc	aaa	aca	agt	ttg	cca	aag	cta	ttg	2320
Leu	Leu	Arg	Ala	Asp	Glu	Asn	Thr	Lys	Thr	Ser	Leu	Pro	Lys	Leu	Leu	
			730				735					740				
ata	gat	gga	cct	tat	gga	gct	cca	gca	caa	gat	tac	cga	aag	tat	gat	2368
Ile	Asp	Gly	Pro	Tyr	Gly	Ala	Pro	Ala	Gln	Asp	Tyr	Arg	Lys	Tyr	Asp	
	745					750					755					
gtc	tta	ctg	ctt	gtt	ggt	ctt	ggc	att	gga	gca	act	ccc	ttt	ata	agt	2416
Val	Leu	Leu	Leu	Val	Gly	Leu	Gly	Ile	Gly	Ala	Thr	Pro	Phe	Ile	Ser	
760					765					770					775	
atc	ctg	aaa	gac	ttg	ctc	aaa	aac	atc	gtc	aca	atg	gag	gag	caa	gca	2464
Ile	Leu	Lys	Asp	Leu	Leu	Lys	Asn	Ile	Val	Thr	Met	Glu	Glu	Gln	Ala	
			780						785					790		
gat	tta	gtc	tcg	gat	ttt	tca	ggg	aac	tca	gac	atg	agc	gct	gca	aca	2512
Asp	Leu	Val	Ser	Asp	Phe	Ser	Gly	Asn	Ser	Asp	Met	Ser	Ala	Ala	Thr	
			795					800					805			

18

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agt gaa caa cca gct ctc aac aag att tct cca aaa aag aga aag agt 2560
Ser Glu Gln Pro Ala Leu Asn Lys Ile Ser Pro Lys Lys Arg Lys Ser
      810                      815                      820

act cta aaa acc aca aat gca tat ttt tat tgg gtg acc cgg gag caa 2608
Thr Leu Lys Thr Thr Asn Ala Tyr Phe Tyr Trp Val Thr Arg Glu Gln
      825                      830                      835

gga tca ttt gat tgg ttc aaa ggt gtt atg aac gaa gtg gct gaa ctt 2656
Gly Ser Phe Asp Trp Phe Lys Gly Val Met Asn Glu Val Ala Glu Leu
840                      845                      850                      855

gat caa agg ggg gtc atc gag atg cat aac tac tta acg agt gtt tat 2704
Asp Gln Arg Gly Val Ile Glu Met His Asn Tyr Leu Thr Ser Val Tyr
      860                      865                      870

gag gaa ggg gat gca cgt tca gct ctc att acc atg gtc cag gcg ctt 2752
Glu Glu Gly Asp Ala Arg Ser Ala Leu Ile Thr Met Val Gln Ala Leu
      875                      880                      885

aac cat gct aag aat ggg gtt gat att gta tca ggc acc agt gtg agg 2800
Asn His Ala Lys Asn Gly Val Asp Ile Val Ser Gly Thr Ser Val Arg
      890                      895                      900

aca cat ttt gcc aga ccg aat tgg agg aaa gta ttt tcc aag acc tta 2848
Thr His Phe Ala Arg Pro Asn Trp Arg Lys Val Phe Ser Lys Thr Leu
      905                      910                      915

acc aag cat gca aat gca aga ata gga gtt ttc tac tgc ggt gca ccc 2896
Thr Lys His Ala Asn Ala Arg Ile Gly Val Phe Tyr Cys Gly Ala Pro
920                      925                      930                      935

ata tta gct aaa gaa ctc agc aaa ctc tgc aaa gag ttt aac caa aag 2944
Ile Leu Ala Lys Glu Leu Ser Lys Leu Cys Lys Glu Phe Asn Gln Lys
      940                      945                      950

ggc aca acg aag ttc gag ttt cac aaa gaa cat ttt tagaaggccc 2990
Gly Thr Thr Lys Phe Glu Phe His Lys Glu His Phe
      955                      960

tggagtacaa ttaatcttgc atcaacggta cacacatcgg taaaccagta tttaccacat 3050
ctatcttttg tacctgattt gatgattcta ctgaagacat aacattagta aggaataagt 3110
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<212> PRT
<213> Solanum tuberosum

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19

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Ser	Ser	Gly	Lys	Asp	Leu	Ser	Gly	Glu	Ser	Ser	Pro	Gly	Thr	Asp	Ser
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Gly	Asn	Ile	Ser	Gly	Phe	Ala	Ser	Glu	Glu	Phe	Val	Glu	Val	Ile	Leu
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Asp	Leu	Gln	Asp	Asp	Asp	Thr	Ile	Ile	Leu	Arg	Ser	Val	Glu	Pro	Ala
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Thr	Val	Ile	Asn	Ile	Asp	Ala	Ser	Asp	Pro	Ala	Thr	Gly	Val	Gly	Ile
	65				70					75					80
Gly	Gly	Val	Ser	Ile	Glu	Thr	Pro	Ala	Ser	Leu	Thr	Ser	Thr	Ser	Gly
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Thr	Arg	Ser	Pro	Thr	Met	Arg	Arg	Ser	Thr	Ser	Asn	Lys	Leu	Arg	Gln
			100					105					110		
Phe	Ser	Gln	Glu	Leu	Lys	Ala	Glu	Ala	Val	Ala	Lys	Ala	Lys	His	Phe
		115					120					125			
Ser	Gln	Glu	Leu	Lys	Ala	Glu	Leu	Arg	Arg	Phe	Ser	Trp	Ser	His	Gly
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His	Ala	Ser	Arg	Thr	Phe	Ser	Pro	Ala	Ser	Phe	Phe	Gln	Asn	Ala	Val
	145				150					155					160
Val	Gly	Thr	Gly	Asn	Gly	Val	Asp	Ser	Ala	Leu	Ala	Ala	Arg	Ala	Leu
				165					170					175	
Arg	Arg	Gln	Arg	Ala	Gln	Leu	Asp	Arg	Thr	Arg	Ser	Ser	Ala	His	Lys
			180					185					190		
Ala	Leu	Arg	Gly	Leu	Lys	Phe	Ile	Ser	Asn	Asn	Lys	Thr	Asn	Gly	Trp
		195					200					205			
Asn	Glu	Val	Glu	Asn	Asn	Phe	Ala	Lys	Leu	Ala	Lys	Asp	Gly	Tyr	Leu
	210					215					220				
Tyr	Arg	Ser	Asp	Phe	Ala	Gln	Cys	Ile	Gly	Met	Lys	Asp	Ser	Lys	Glu
	225				230					235					240
Phe	Ala	Leu	Glu	Leu	Phe	Asp	Ala	Leu	Ser	Arg	Arg	Arg	Arg	Leu	Lys
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Val	Asp	Lys	Ile	Ser	Lys	Glu	Glu	Leu	Tyr	Glu	Tyr	Trp	Ser	Gln	Ile
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Thr	Asp	Gln	Ser	Phe	Asp	Ser	Arg	Leu	Gln	Ile	Phe	Phe	Asp	Met	Val
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Asp	Lys	Asn	Glu	Asp	Gly	Arg	Ile	Gly	Glu	Glu	Glu	Val	Lys	Glu	Ile
	290					295					300				
Ile	Met	Leu	Ser	Ala	Ser	Ala	Asn	Lys	Leu	Ser	Arg	Leu	Lys	Glu	Gln
	305				310					315					320
Ala	Glu	Glu	Tyr	Ala	Ala	Leu	Ile	Met	Glu	Glu	Leu	Asp	Pro	Glu	Arg
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Leu	Gly	Tyr	Ile	Glu	Leu	Trp	Gln	Leu	Glu	Thr	Leu	Leu	Leu	Gln	Lys
			340					345					350		
Asp	Thr	Tyr	Leu	Asn	Tyr	Ser	Gln	Ala	Leu	Ser	Tyr	Thr	Ser	Gln	Ala
		355					360					365			

20

Leu	Ser	Gln	Asn	Leu	Gln	Gly	Leu	Arg	Lys	Arg	Ser	Pro	Ile	Arg	Arg	370	375	380	
Met	Ser	Thr	Lys	Leu	Val	Tyr	Ser	Leu	Gln	Glu	Asn	Trp	Lys	Arg	Ile	385	390	395	400
Trp	Val	Leu	Val	Leu	Trp	Ile	Leu	Ile	Met	Ile	Gly	Leu	Phe	Leu	Trp	405	410	415	
Lys	Phe	Tyr	Leu	Tyr	Lys	Gln	Lys	Ser	Ala	Phe	Gln	Val	Met	Gly	Tyr	420	425	430	
Cys	Leu	Leu	Thr	Ala	Lys	Gly	Ala	Ala	Glu	Thr	Leu	Lys	Phe	Asn	Met	435	440	445	
Ala	Leu	Ile	Leu	Leu	Pro	Val	Cys	Arg	Asn	Thr	Ile	Thr	Phe	Leu	Arg	450	455	460	
Ser	Thr	Lys	Leu	Ser	Cys	Phe	Val	Pro	Phe	Asp	Asp	Asn	Ile	Asn	Phe	465	470	475	480
His	Lys	Thr	Val	Ala	Ala	Ala	Ile	Val	Thr	Gly	Ile	Ile	Leu	His	Ala	485	490	495	
Gly	Asn	His	Leu	Val	Cys	Asp	Phe	Pro	Lys	Leu	Ile	His	Ala	Asn	Asn	500	505	510	
Thr	Asn	Tyr	Gln	Lys	Tyr	Leu	Val	Asn	Asp	Phe	Gly	Pro	Ser	Gln	Pro	515	520	525	
Gln	Tyr	Ile	Asp	Leu	Val	Lys	Gly	Val	Glu	Gly	Val	Thr	Gly	Ile	Ile	530	535	540	
Met	Val	Ile	Leu	Met	Ala	Ile	Ala	Phe	Thr	Leu	Ala	Thr	Arg	Trp	Phe	545	550	555	560
Arg	Arg	Ser	Leu	Ile	Lys	Phe	Pro	Lys	Pro	Phe	Asp	Arg	Leu	Thr	Gly	565	570	575	
Phe	Asn	Ala	Phe	Trp	Tyr	Ser	His	His	Leu	Leu	Ile	Ile	Val	Tyr	Ile	580	585	590	
Val	Leu	Ile	Ile	His	Gly	Thr	Phe	Leu	Tyr	Leu	Val	His	Asn	Trp	Tyr	595	600	605	
Ser	Lys	Thr	Thr	Trp	Met	Tyr	Leu	Ala	Val	Pro	Val	Leu	Leu	Tyr	Ala	610	615	620	
Gly	Glu	Arg	Thr	Leu	Arg	Phe	Phe	Arg	Ser	Gly	Leu	Tyr	Thr	Val	Arg	625	630	635	640
Leu	Leu	Lys	Val	Ala	Ile	Tyr	Pro	Gly	Asn	Val	Leu	Thr	Leu	Gln	Met	645	650	655	
Ser	Lys	Pro	Pro	Gln	Phe	Arg	Tyr	Lys	Ser	Gly	Gln	Tyr	Met	Phe	Val	660	665	670	
Gln	Cys	Pro	Ala	Val	Ser	Pro	Phe	Glu	Trp	His	Pro	Phe	Ser	Ile	Thr	675	680	685	
Ser	Ala	Pro	Gly	Asp	Asp	Tyr	Leu	Ser	Ile	His	Ile	Arg	Gln	Leu	Gly	690	695	700	
Asp	Trp	Thr	Gln	Glu	Leu	Lys	Arg	Val	Phe	Ser	Glu	Ala	Cys	Glu	Gln	705	710	715	720
Pro	Glu	Ala	Gly	Lys	Ser	Gly	Leu	Leu	Arg	Ala	Asp	Glu	Asn	Thr	Lys	725	730	735	
Thr	Ser	Leu	Pro	Lys	Leu	Leu	Ile	Asp	Gly	Pro	Tyr	Gly	Ala	Pro	Ala	740	745	750	

21

Gln Asp Tyr Arg Lys Tyr Asp Val Leu Leu Leu Val Gly Leu Gly Ile
 755 760 765
 Gly Ala Thr Pro Phe Ile Ser Ile Leu Lys Asp Leu Leu Lys Asn Ile
 770 775 780
 Val Thr Met Glu Glu Gln Ala Asp Leu Val Ser Asp Phe Ser Gly Asn
 785 790 795 800
 Ser Asp Met Ser Ala Ala Thr Ser Glu Gln Pro Ala Leu Asn Lys Ile
 805 810 815
 Ser Pro Lys Lys Arg Lys Ser Thr Leu Lys Thr Thr Asn Ala Tyr Phe
 820 825 830
 Tyr Trp Val Thr Arg Glu Gln Gly Ser Phe Asp Trp Phe Lys Gly Val
 835 840 845
 Met Asn Glu Val Ala Glu Leu Asp Gln Arg Gly Val Ile Glu Met His
 850 855 860
 Asn Tyr Leu Thr Ser Val Tyr Glu Glu Gly Asp Ala Arg Ser Ala Leu
 865 870 875 880
 Ile Thr Met Val Gln Ala Leu Asn His Ala Lys Asn Gly Val Asp Ile
 885 890 895
 Val Ser Gly Thr Ser Val Arg Thr His Phe Ala Arg Pro Asn Trp Arg
 900 905 910
 Lys Val Phe Ser Lys Thr Leu Thr Lys His Ala Asn Ala Arg Ile Gly
 915 920 925
 Val Phe Tyr Cys Gly Ala Pro Ile Leu Ala Lys Glu Leu Ser Lys Leu
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 Cys Lys Glu Phe Asn Gln Lys Gly Thr Thr Lys Phe Glu Phe His Lys
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<212> DNA

<213> Lycopersicon esculentum

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<221> CDS

<222> (146)..(3112)

<223> coding for NADPH oxidase

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 tttattgtca tttgatttgg gacag atg agg ggt tta cct ggg cat gaa cgc 172
 Met Arg Gly Leu Pro Gly His Glu Arg
 1 5
 cgg tgg acg tcg gat acg gtg tct tcc ggg aag gat tta agt ggt gag 220
 Arg Trp Thr Ser Asp Thr Val Ser Ser Gly Lys Asp Leu Ser Gly Glu
 10 15 20 25
 tca tcg ccg gga act gat tcc ggg aat att tcc ggt ttt gct tcg gag 268
 Ser Ser Pro Gly Thr Asp Ser Gly Asn Ile Ser Gly Phe Ala Ser Glu
 30 35 40

22

gag ttt gtt gaa gtt ata ctt gat ctt cag gat gat gat acg att att	316
Glu Phe Val Glu Val Ile Leu Asp Leu Gln Asp Asp Asp Thr Ile Ile	
45 50 55	
tta cgg agc gtt gaa ccg gct act gta atc aac att gat ggt tct gat	364
Leu Arg Ser Val Glu Pro Ala Thr Val Ile Asn Ile Asp Gly Ser Asp	
60 65 70	
cct gct tcc gga gtc ggt att ggt gga gca tcg att gaa act ccg gcg	412
Pro Ala Ser Gly Val Gly Ile Gly Gly Ala Ser Ile Glu Thr Pro Ala	
75 80 85	
tcg gtg acg tcg acg tcg gaa act cga tcg ccg atg atg cgt cgg agt	460
Ser Val Thr Ser Thr Ser Glu Thr Arg Ser Pro Met Met Arg Arg Ser	
90 95 100 105	
aca tct aat aag ttt cgt cag ttt tca cag gag ttg aaa gct gag gct	508
Thr Ser Asn Lys Phe Arg Gln Phe Ser Gln Glu Leu Lys Ala Glu Ala	
110 115 120	
gtt gcg aaa gcg aag cat ttc tcg caa gag ctt aaa gcg gag cta agg	556
Val Ala Lys Ala Lys His Phe Ser Gln Glu Leu Lys Ala Glu Leu Arg	
125 130 135	
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Arg Phe Ser Trp Ser His Gly His Ala Ser Arg Ala Phe Ser Pro Ala	
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tcg ttt ttc caa aac gct gtc gtc gga aca ggc aac ggt gta gac tcg	652
Ser Phe Phe Gln Asn Ala Val Val Gly Thr Gly Asn Gly Val Asp Ser	
155 160 165	
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Ala Leu Ala Ala Arg Ala Leu Arg Arg Gln Arg Ala Gln Leu Asp Arg	
170 175 180 185	
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Thr Arg Ser Ser Ala His Lys Ala Leu Arg Gly Leu Lys Phe Ile Ser	
190 195 200	
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Asn Asn Lys Thr Asn Gly Trp Asn Glu Val Glu Asn Asn Phe Ala Lys	
205 210 215	
ctc gct aaa gac ggt tac ctt tat cgt tcc gat ttc gca caa tgc atc	844
Leu Ala Lys Asp Gly Tyr Leu Tyr Arg Ser Asp Phe Ala Gln Cys Ile	
220 225 230	
ggt cag tac tca cgc ccg cga tca cta cag ttt aat tat aga tta att	892
Gly Gln Tyr Ser Arg Arg Arg Ser Leu Gln Phe Asn Tyr Arg Leu Ile	
235 240 245	
aca tta att ttg att aat tat ttg gtt aaa ggt atg aag gat tca aag	940
Thr Leu Ile Leu Ile Asn Tyr Leu Val Lys Gly Met Lys Asp Ser Lys	
250 255 260 265	
gaa ttt gcg ttg gaa ttg ttt gat gct tta agt aga aga aga ttg	988
Glu Phe Ala Leu Glu Leu Phe Asp Ala Leu Ser Arg Arg Arg Arg Leu	
270 275 280	
aag gtt gat aag att agc caa gag gaa ttg tat gag tat tgg tct caa	1036
Lys Val Asp Lys Ile Ser Gln Glu Glu Leu Tyr Glu Tyr Trp Ser Gln	
285 290 295	
atc acc gat cag agt ttc gat tct ccg ctt cag atc ttc ttc gac atg	1084
Ile Thr Asp Gln Ser Phe Asp Ser Arg Leu Gln Ile Phe Phe Asp Met	
300 305 310	

23

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Val Asp Lys Asn Glu Asp Gly Arg Ile Gly Glu Glu Glu Val Lys Glu	
315 320 325	
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Ile Ile Met Leu Ser Ala Ser Ala Asn Lys Leu Ser Arg Leu Lys Glu	
330 335 340 345	
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Gln Ala Glu Glu Tyr Ala Ala Leu Ile Met Glu Glu Leu Asp Pro Glu	
350 355 360	
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Arg Leu Gly Tyr Ile Glu Leu Trp Gln Leu Glu Thr Leu Leu Leu Gln	
365 370 375	
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Lys Asp Thr Tyr Leu Asn Tyr Ser Gln Ala Leu Ser Tyr Thr Ser Gln	
380 385 390	
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Ala Leu Ser Gln Asn Leu Gln Gly Leu Arg Lys Arg Ser Pro Ile Arg	
395 400 405	
aga atg agc aca aaa ctt gtc tat tca ctg caa gag aat tgg aag aga	1420
Arg Met Ser Thr Lys Leu Val Tyr Ser Leu Gln Glu Asn Trp Lys Arg	
410 415 420 425	
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Ile Trp Val Leu Val Leu Trp Ile Leu Ile Met Ile Gly Leu Phe Leu	
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Trp Lys Phe Tyr Gln Tyr Lys Gln Lys Ser Ala Phe Gln Val Met Gly	
445 450 455	
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Tyr Cys Leu Leu Thr Ala Lys Gly Ala Ala Glu Thr Leu Lys Phe Asn	
460 465 470	
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Met Ala Leu Ile Leu Leu Pro Val Cys Arg Asn Thr Ile Thr Phe Leu	
475 480 485	
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Arg Ser Thr Lys Leu Ser Cys Phe Val Pro Phe Asp Asp Asn Ile Asn	
490 495 500 505	
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Phe His Lys Thr Val Ala Ala Ala Ile Val Thr Gly Ile Ile Leu His	
510 515 520	
gcc ggt aat cac ctt gta tgt gat ttc cca aag ctt ata cat gca aat	1756
Ala Gly Asn His Leu Val Cys Asp Phe Pro Lys Leu Ile His Ala Asn	
525 530 535	
agt acg aat tat cag aaa tat ttg gtg aat gat ttt ggc cca agc cag	1804
Ser Thr Asn Tyr Gln Lys Tyr Leu Val Asn Asp Phe Gly Pro Ser Gln	
540 545 550	
cct cag tac ata gat ctt gtt aaa gga gtg gag ggt gtg act gga ata	1852
Pro Gln Tyr Ile Asp Leu Val Lys Gly Val Glu Gly Val Thr Gly Ile	
555 560 565	
gtt atg gta atc ctc atg gcc att gct ttc act ctt gca acg cga tgg	1900
Val Met Val Ile Leu Met Ala Ile Ala Phe Thr Leu Ala Thr Arg Trp	
570 575 580 585	

24

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Phe Arg Arg Ser Leu Ile Lys Leu Pro Lys Pro Phe Asp Arg Leu Thr	
590 595 600	
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Gly Phe Asn Ala Phe Trp Tyr Ser His His Leu Leu Ile Ile Val Tyr	
605 610 615	
atc gta ctg atc atc cat ggc aca ttc ctc tac ctt gtg cat aac tgg	2044
Ile Val Leu Ile Ile His Gly Thr Phe Leu Tyr Leu Val His Asn Trp	
620 625 630	
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Tyr Ser Lys Thr Thr Trp Met Tyr Ile Ala Val Pro Val Leu Leu Tyr	
635 640 645	
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Ala Gly Glu Arg Thr Leu Arg Phe Phe Arg Ser Gly Leu Tyr Ser Val	
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cgg ctt cta aaa gta gca ata tat cct gga aat gtc ctt act ctg caa	2188
Arg Leu Leu Lys Val Ala Ile Tyr Pro Gly Asn Val Leu Thr Leu Gln	
670 675 680	
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Met Ser Lys Pro Pro Gln Phe Arg Tyr Lys Ser Gly Gln Tyr Met Phe	
685 690 695	
gtc cag tgt cca gct gtt tct cca ttc gag tgg cat cca ttt tcc att	2284
Val Gln Cys Pro Ala Val Ser Pro Phe Glu Trp His Pro Phe Ser Ile	
700 705 710	
act tca gct cct ggg gat gac tac ttg agc att cat atc cga caa ctt	2332
Thr Ser Ala Pro Gly Asp Asp Tyr Leu Ser Ile His Ile Arg Gln Leu	
715 720 725	
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Gly Asp Trp Thr Gln Glu Leu Lys Arg Val Phe Ser Glu Ala Cys Glu	
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Gln Pro Glu Ala Gly Lys Ser Gly Leu Leu Arg Ala Asp Glu Asn Thr	
750 755 760	
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Lys Thr Ser Leu Pro Lys Leu Leu Ile Asp Gly Pro Tyr Gly Ala Pro	
765 770 775	
gca caa gat tac cgg aag tat gat gtc tta ctg ctt gtt ggt ctt ggc	2524
Ala Gln Asp Tyr Arg Lys Tyr Asp Val Leu Leu Leu Val Gly Leu Gly	
780 785 790	
att gga gca act ccc ttt ata agt atc ctg aaa gac ttg ctc aaa aac	2572
Ile Gly Ala Thr Pro Phe Ile Ser Ile Leu Lys Asp Leu Leu Lys Asn	
795 800 805	
atc gtc gca atg gag gag caa gca gat tta gtc tcg gat ttc agt gga	2620
Ile Val Ala Met Glu Glu Gln Ala Asp Leu Val Ser Asp Phe Ser Gly	
810 815 820 825	
aac tcg gac atg agt gct gca aca agt gaa caa cca gct ctc aac aag	2668
Asn Ser Asp Met Ser Ala Ala Thr Ser Glu Gln Pro Ala Leu Asn Lys	
830 835 840	
att tct cca aaa aag aga aag agt act cta aaa acc aca aat gca tat	2716
Ile Ser Pro Lys Lys Arg Lys Ser Thr Leu Lys Thr Thr Asn Ala Tyr	
845 850 855	

25

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ttt tat tgg gtg acc cgg gag caa gga tca ttt gat tgg ttc aaa ggt 2764
Phe Tyr Trp Val Thr Arg Glu Gln Gly Ser Phe Asp Trp Phe Lys Gly
      860                      865                      870

gtt atg aat gaa gtg gct gaa ctt gat caa agg ggt gtc atc gag atg 2812
Val Met Asn Glu Val Ala Glu Leu Asp Gln Arg Gly Val Ile Glu Met
      875                      880                      885

cat aac tac ttg acg agt gtt tat gag gaa ggg gat gca cgt tca gct 2860
His Asn Tyr Leu Thr Ser Val Tyr Glu Glu Gly Asp Ala Arg Ser Ala
      890                      895                      900                      905

ctc att acc atg gtc cag gca ctt aac cat gct aag aat ggg gtt gat 2908
Leu Ile Thr Met Val Gln Ala Leu Asn His Ala Lys Asn Gly Val Asp
      910                      915                      920

att gta tca ggc acc agt gtg agg aca cat ttc gcc agg ccg aat tgg 2956
Ile Val Ser Gly Thr Ser Val Arg Thr His Phe Ala Arg Pro Asn Trp
      925                      930                      935

agg aaa gta ttt tcc aag acc tta acc aag cat gca aat gca aga ata 3004
Arg Lys Val Phe Ser Lys Thr Leu Thr Lys His Ala Asn Ala Arg Ile
      940                      945                      950

gga gtt ttc tac tgt ggt gca ccc ata tta gct aaa gaa ctc agc caa 3052
Gly Val Phe Tyr Cys Gly Ala Pro Ile Leu Ala Lys Glu Leu Ser Gln
      955                      960                      965

ctc tgc aaa gag ttt aac caa aag ggc aca aca aag ttc gag ttt cac 3100
Leu Cys Lys Glu Phe Asn Gln Lys Gly Thr Thr Lys Phe Glu Phe His
      970                      975                      980                      985

aaa gaa cat ttt tagaagggcc tggagtatga ttaatcttgcc atcaacggta 3152
Lys Glu His Phe

cacacatcta tcttcggtac cttatttgat tattctactg aagagataac attagtaagg 3212
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Gly Asn Ile Ser Gly Phe Ala Ser Glu Glu Phe Val Glu Val Ile Leu
      35                      40                      45
Asp Leu Gln Asp Asp Asp Thr Ile Ile Leu Arg Ser Val Glu Pro Ala
      50                      55                      60
Thr Val Ile Asn Ile Asp Gly Ser Asp Pro Ala Ser Gly Val Gly Ile
      65                      70                      75                      80
Gly Gly Ala Ser Ile Glu Thr Pro Ala Ser Val Thr Ser Thr Ser Glu
      85                      90                      95
Thr Arg Ser Pro Met Met Arg Arg Ser Thr Ser Asn Lys Phe Arg Gln
      100                      105                      110
Phe Ser Gln Glu Leu Lys Ala Glu Ala Val Ala Lys Ala Lys His Phe
      115                      120                      125

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Ser	Gln	Glu	Leu	Lys	Ala	Glu	Leu	Arg	Arg	Phe	Ser	Trp	Ser	His	Gly	130	135	140
His	Ala	Ser	Arg	Ala	Phe	Ser	Pro	Ala	Ser	Phe	Phe	Gln	Asn	Ala	Val	145	150	155
Val	Gly	Thr	Gly	Asn	Gly	Val	Asp	Ser	Ala	Leu	Ala	Ala	Arg	Ala	Leu	165	170	175
Arg	Arg	Gln	Arg	Ala	Gln	Leu	Asp	Arg	Thr	Arg	Ser	Ser	Ala	His	Lys	180	185	190
Ala	Leu	Arg	Gly	Leu	Lys	Phe	Ile	Ser	Asn	Asn	Lys	Thr	Asn	Gly	Trp	195	200	205
Asn	Glu	Val	Glu	Asn	Asn	Phe	Ala	Lys	Leu	Ala	Lys	Asp	Gly	Tyr	Leu	210	215	220
Tyr	Arg	Ser	Asp	Phe	Ala	Gln	Cys	Ile	Gly	Gln	Tyr	Ser	Arg	Arg	Arg	225	230	235
Ser	Leu	Gln	Phe	Asn	Tyr	Arg	Leu	Ile	Thr	Leu	Ile	Leu	Ile	Asn	Tyr	245	250	255
Leu	Val	Lys	Gly	Met	Lys	Asp	Ser	Lys	Glu	Phe	Ala	Leu	Glu	Leu	Phe	260	265	270
Asp	Ala	Leu	Ser	Arg	Arg	Arg	Arg	Leu	Lys	Val	Asp	Lys	Ile	Ser	Gln	275	280	285
Glu	Glu	Leu	Tyr	Glu	Tyr	Trp	Ser	Gln	Ile	Thr	Asp	Gln	Ser	Phe	Asp	290	295	300
Ser	Arg	Leu	Gln	Ile	Phe	Phe	Asp	Met	Val	Asp	Lys	Asn	Glu	Asp	Gly	305	310	315
Arg	Ile	Gly	Glu	Glu	Glu	Val	Lys	Glu	Ile	Ile	Met	Leu	Ser	Ala	Ser	325	330	335
Ala	Asn	Lys	Leu	Ser	Arg	Leu	Lys	Glu	Gln	Ala	Glu	Glu	Tyr	Ala	Ala	340	345	350
Leu	Ile	Met	Glu	Glu	Leu	Asp	Pro	Glu	Arg	Leu	Gly	Tyr	Ile	Glu	Leu	355	360	365
Trp	Gln	Leu	Glu	Thr	Leu	Leu	Leu	Gln	Lys	Asp	Thr	Tyr	Leu	Asn	Tyr	370	375	380
Ser	Gln	Ala	Leu	Ser	Tyr	Thr	Ser	Gln	Ala	Leu	Ser	Gln	Asn	Leu	Gln	385	390	395
Gly	Leu	Arg	Lys	Arg	Ser	Pro	Ile	Arg	Arg	Met	Ser	Thr	Lys	Leu	Val	405	410	415
Tyr	Ser	Leu	Gln	Glu	Asn	Trp	Lys	Arg	Ile	Trp	Val	Leu	Val	Leu	Trp	420	425	430
Ile	Leu	Ile	Met	Ile	Gly	Leu	Phe	Leu	Trp	Lys	Phe	Tyr	Gln	Tyr	Lys	435	440	445
Gln	Lys	Ser	Ala	Phe	Gln	Val	Met	Gly	Tyr	Cys	Leu	Leu	Thr	Ala	Lys	450	455	460
Gly	Ala	Ala	Glu	Thr	Leu	Lys	Phe	Asn	Met	Ala	Leu	Ile	Leu	Leu	Pro	465	470	475
Val	Cys	Arg	Asn	Thr	Ile	Thr	Phe	Leu	Arg	Ser	Thr	Lys	Leu	Ser	Cys	485	490	495
Phe	Val	Pro	Phe	Asp	Asp	Asn	Ile	Asn	Phe	His	Lys	Thr	Val	Ala	Ala	500	505	510

27

Ala	Ile	Val	Thr	Gly	Ile	Ile	Leu	His	Ala	Gly	Asn	His	Leu	Val	Cys
		515					520					525			
Asp	Phe	Pro	Lys	Leu	Ile	His	Ala	Asn	Ser	Thr	Asn	Tyr	Gln	Lys	Tyr
	530					535					540				
Leu	Val	Asn	Asp	Phe	Gly	Pro	Ser	Gln	Pro	Gln	Tyr	Ile	Asp	Leu	Val
545					550					555					560
Lys	Gly	Val	Glu	Gly	Val	Thr	Gly	Ile	Val	Met	Val	Ile	Leu	Met	Ala
				565					570					575	
Ile	Ala	Phe	Thr	Leu	Ala	Thr	Arg	Trp	Phe	Arg	Arg	Ser	Leu	Ile	Lys
			580					585					590		
Leu	Pro	Lys	Pro	Phe	Asp	Arg	Leu	Thr	Gly	Phe	Asn	Ala	Phe	Trp	Tyr
		595					600					605			
Ser	His	His	Leu	Leu	Ile	Ile	Val	Tyr	Ile	Val	Leu	Ile	Ile	His	Gly
	610					615					620				
Thr	Phe	Leu	Tyr	Leu	Val	His	Asn	Trp	Tyr	Ser	Lys	Thr	Thr	Trp	Met
625					630					635					640
Tyr	Ile	Ala	Val	Pro	Val	Leu	Leu	Tyr	Ala	Gly	Glu	Arg	Thr	Leu	Arg
				645					650					655	
Phe	Phe	Arg	Ser	Gly	Leu	Tyr	Ser	Val	Arg	Leu	Leu	Lys	Val	Ala	Ile
			660					665					670		
Tyr	Pro	Gly	Asn	Val	Leu	Thr	Leu	Gln	Met	Ser	Lys	Pro	Pro	Gln	Phe
		675					680					685			
Arg	Tyr	Lys	Ser	Gly	Gln	Tyr	Met	Phe	Val	Gln	Cys	Pro	Ala	Val	Ser
	690					695					700				
Pro	Phe	Glu	Trp	His	Pro	Phe	Ser	Ile	Thr	Ser	Ala	Pro	Gly	Asp	Asp
705					710					715					720
Tyr	Leu	Ser	Ile	His	Ile	Arg	Gln	Leu	Gly	Asp	Trp	Thr	Gln	Glu	Leu
				725					730					735	
Lys	Arg	Val	Phe	Ser	Glu	Ala	Cys	Glu	Gln	Pro	Glu	Ala	Gly	Lys	Ser
			740					745					750		
Gly	Leu	Leu	Arg	Ala	Asp	Glu	Asn	Thr	Lys	Thr	Ser	Leu	Pro	Lys	Leu
		755					760					765			
Leu	Ile	Asp	Gly	Pro	Tyr	Gly	Ala	Pro	Ala	Gln	Asp	Tyr	Arg	Lys	Tyr
	770					775					780				
Asp	Val	Leu	Leu	Leu	Val	Gly	Leu	Gly	Ile	Gly	Ala	Thr	Pro	Phe	Ile
785					790					795					800
Ser	Ile	Leu	Lys	Asp	Leu	Leu	Lys	Asn	Ile	Val	Ala	Met	Glu	Glu	Gln
				805					810					815	
Ala	Asp	Leu	Val	Ser	Asp	Phe	Ser	Gly	Asn	Ser	Asp	Met	Ser	Ala	Ala
			820					825					830		
Thr	Ser	Glu	Gln	Pro	Ala	Leu	Asn	Lys	Ile	Ser	Pro	Lys	Lys	Arg	Lys
		835					840					845			
Ser	Thr	Leu	Lys	Thr	Thr	Asn	Ala	Tyr	Phe	Tyr	Trp	Val	Thr	Arg	Glu
	850					855					860				
Gln	Gly	Ser	Phe	Asp	Trp	Phe	Lys	Gly	Val	Met	Asn	Glu	Val	Ala	Glu
865					870					875					880
Leu	Asp	Gln	Arg	Gly	Val	Ile	Glu	Met	His	Asn	Tyr	Leu	Thr	Ser	Val
				885					890						895

28

Tyr Glu Glu Gly Asp Ala Arg Ser Ala Leu Ile Thr Met Val Gln Ala
 900 905 910
 Leu Asn His Ala Lys Asn Gly Val Asp Ile Val Ser Gly Thr Ser Val
 915 920 925
 Arg Thr His Phe Ala Arg Pro Asn Trp Arg Lys Val Phe Ser Lys Thr
 930 935 940
 Leu Thr Lys His Ala Asn Ala Arg Ile Gly Val Phe Tyr Cys Gly Ala
 945 950 955 960
 Pro Ile Leu Ala Lys Glu Leu Ser Gln Leu Cys Lys Glu Phe Asn Gln
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<222> (15)..(2846)

<223> coding for NADPH oxidase

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 1 5 10
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 Ser Phe Asp Ser Val Ser Ala Gly Lys Thr Ala Val Gly Ser Ala Ser
 15 20 25
 act tca ccg gga act gaa tac tcc att aac ggt gat caa gag ttc gtt 146
 Thr Ser Pro Gly Thr Glu Tyr Ser Ile Asn Gly Asp Gln Glu Phe Val
 30 35 40
 gaa gtc aca atc gat ctt caa gac gat gac aca atc gtt ctt cgt agc 194
 Glu Val Thr Ile Asp Leu Gln Asp Asp Asp Thr Ile Val Leu Arg Ser
 45 50 55 60
 gtc gag cca gca acc gcc att aat gtc atc gga gat atc tcc gac gac 242
 Val Glu Pro Ala Thr Ala Ile Asn Val Ile Gly Asp Ile Ser Asp Asp
 65 70 75
 aac acc gga ata atg act ccg gtt tcg att tcg aga tct ccg acg atg 290
 Asn Thr Gly Ile Met Thr Pro Val Ser Ile Ser Arg Ser Pro Thr Met
 80 85 90
 aaa cga act tca tct aat ccg ttc cga caa ttc tca caa gag ctt aaa 338
 Lys Arg Thr Ser Ser Asn Arg Phe Arg Gln Phe Ser Gln Glu Leu Lys
 95 100 105
 gcc gaa gct gtg gcg aaa gcg aaa cag tta tct cag gag ttg aaa cga 386
 Ala Glu Ala Val Ala Lys Ala Lys Gln Leu Ser Gln Glu Leu Lys Arg
 110 115 120
 ttc tca tgg tct cgt tct ttc tca ggt aac tta acc act act agt acc 434
 Phe Ser Trp Ser Arg Ser Phe Ser Gly Asn Leu Thr Thr Thr Ser Thr
 125 130 135 140
 gcc gct aat caa agc ggc ggt gct ggt ggt ggt ttg gtg aac tcg gct 482
 Ala Ala Asn Gln Ser Gly Gly Ala Gly Gly Gly Leu Val Asn Ser Ala
 145 150 155

29

tta gaa gcg cga gcg ttg cga aag caa cgt gct cag tta gat cgg act	530
Leu Glu Ala Arg Ala Leu Arg Lys Gln Arg Ala Gln Leu Asp Arg Thr	
160 165 170	
cgg tct agt gct caa aga gct ctt cgt ggt ttg aga ttc att agc aat	578
Arg Ser Ser Ala Gln Arg Ala Leu Arg Gly Leu Arg Phe Ile Ser Asn	
175 180 185	
aag caa aag aac gtt gat ggt tgg aac gat gtt caa tca aat ttc gaa	626
Lys Gln Lys Asn Val Asp Gly Trp Asn Asp Val Gln Ser Asn Phe Glu	
190 195 200	
aaa ttc gaa aaa aat ggt tac atc tat cgc tcc gat ttc gct caa tgc	674
Lys Phe Glu Lys Asn Gly Tyr Ile Tyr Arg Ser Asp Phe Ala Gln Cys	
205 210 215 220	
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Ile Gly Met Lys Asp Ser Lys Glu Phe Ala Leu Glu Leu Phe Asp Ala	
225 230 235	
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Leu Ser Arg Arg Arg Arg Leu Lys Val Glu Lys Ile Asn His Asp Glu	
240 245 250	
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Leu Tyr Glu Tyr Trp Ser Gln Ile Asn Asp Glu Ser Phe Asp Ser Arg	
255 260 265	
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Leu Gln Ile Phe Phe Asp Ile Val Asp Lys Asn Glu Asp Gly Arg Ile	
270 275 280	
aca gaa gag gaa gta aaa gag ata ata atg ttg agt gca tct gca aat	914
Thr Glu Glu Glu Val Lys Glu Ile Ile Met Leu Ser Ala Ser Ala Asn	
285 290 295 300	
aag cta tca aga tta aag gaa caa gca gag gaa tat gca gct ttg att	962
Lys Leu Ser Arg Leu Lys Glu Gln Ala Glu Glu Tyr Ala Ala Leu Ile	
305 310 315	
atg gaa gag tta gat cct gaa aga ctt ggc tac ata gag cta tgg caa	1010
Met Glu Glu Leu Asp Pro Glu Arg Leu Gly Tyr Ile Glu Leu Trp Gln	
320 325 330	
cta gag act ttg ctt cta caa aaa gac aca tac ctc aat tac agt caa	1058
Leu Glu Thr Leu Leu Leu Gln Lys Asp Thr Tyr Leu Asn Tyr Ser Gln	
335 340 345	
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Ala Leu Ser Tyr Thr Ser Gln Ala Leu Ser Gln Asn Leu Gln Gly Leu	
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Arg Gly Lys Ser Arg Ile His Arg Met Ser Ser Asp Phe Val Tyr Ile	
365 370 375 380	
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Met Gln Glu Asn Trp Lys Arg Ile Trp Val Leu Ser Leu Trp Ile Met	
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415 420 425	

30

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Arg Asn Thr Ile Thr Trp Leu Arg Ser Thr Arg Leu Ser Tyr Phe Val	
445 450 455 460	
cct ttt gat gat aat atc aac ttc cac aag aca att gct gga gcc att	1442
Pro Phe Asp Asp Asn Ile Asn Phe His Lys Thr Ile Ala Gly Ala Ile	
465 470 475	
gta gta gct gtg atc ctt cat att gga gac cat ctt gct tgt gat ttc	1490
Val Val Ala Val Ile Leu His Ile Gly Asp His Leu Ala Cys Asp Phe	
480 485 490	
cct aga att gtt aga gcc acc gaa tac gat tac aat cgg tat ctg ttt	1538
Pro Arg Ile Val Arg Ala Thr Glu Tyr Asp Tyr Asn Arg Tyr Leu Phe	
495 500 505	
cat tac ttt caa aca aaa cag cca aca tac ttc gac ctc gtt aag gga	1586
His Tyr Phe Gln Thr Lys Gln Pro Thr Tyr Phe Asp Leu Val Lys Gly	
510 515 520	
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Pro Glu Gly Ile Thr Gly Ile Leu Met Val Ile Leu Met Ile Ile Ser	
525 530 535 540	
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Phe Thr Leu Ala Thr Arg Trp Phe Arg Arg Asn Leu Val Lys Leu Pro	
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Lys Pro Phe Asp Arg Leu Thr Gly Phe Asn Ala Phe Trp Tyr Ser His	
560 565 570	
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His Leu Phe Val Ile Val Tyr Ile Leu Leu Ile Leu His Gly Ile Phe	
575 580 585	
ctc tat ttc gcc aag cct tgg tat gtt cgt acg aca tgg atg tat ctt	1826
Leu Tyr Phe Ala Lys Pro Trp Tyr Val Arg Thr Thr Trp Met Tyr Leu	
590 595 600	
gca gta cca gtt tta ctc tat ggt gga gaa aga aca ctt agg tac ttc	1874
Ala Val Pro Val Leu Leu Tyr Gly Gly Glu Arg Thr Leu Arg Tyr Phe	
605 610 615 620	
cgt tct ggt tct tat tcg gtt cga ctg ctt aag gtt gct ata tat cct	1922
Arg Ser Gly Ser Tyr Ser Val Arg Leu Leu Lys Val Ala Ile Tyr Pro	
625 630 635	
ggt aat gtt cta acg cta caa atg tcg aaa cca act caa ttt cgt tac	1970
Gly Asn Val Leu Thr Leu Gln Met Ser Lys Pro Thr Gln Phe Arg Tyr	
640 645 650	
aaa agc gga caa tac atg ttt gtc caa tgt cct gcg gtt tcg cca ttc	2018
Lys Ser Gly Gln Tyr Met Phe Val Gln Cys Pro Ala Val Ser Pro Phe	
655 660 665	
gag tgg cat cca ttc tca att act tcc gca cct gaa gat gat tat atc	2066
Glu Trp His Pro Phe Ser Ile Thr Ser Ala Pro Glu Asp Asp Tyr Ile	
670 675 680	
agc att cac att aga caa ctt ggt gat tgg act caa gaa ctc aaa aga	2114
Ser Ile His Ile Arg Gln Leu Gly Asp Trp Thr Gln Glu Leu Lys Arg	
685 690 695 700	

31

gta ttc tct gaa gtt tgt gag cca ccg gtt ggc ggt aaa agc gga ctt	2162
Val Phe Ser Glu Val Cys Glu Pro Pro Val Gly Gly Lys Ser Gly Leu	
705 710 715	
ctc aga gcc gac gaa aca aca aag aaa agt ttg cca aag cta ttg ata	2210
Leu Arg Ala Asp Glu Thr Thr Lys Lys Ser Leu Pro Lys Leu Leu Ile	
720 725 730	
gat gga ccg tac ggt gca cca gca caa gat tat agg aaa tat gat gtt	2258
Asp Gly Pro Tyr Gly Ala Pro Ala Gln Asp Tyr Arg Lys Tyr Asp Val	
735 740 745	
ctc tta tta gtt ggt ctt ggc att ggt gca act cca ttt atc agt atc	2306
Leu Leu Leu Val Gly Leu Gly Ile Gly Ala Thr Pro Phe Ile Ser Ile	
750 755 760	
ttg aaa gat ttg ctt aac aac att gtt aaa atg gaa gag cat gcg gat	2354
Leu Lys Asp Leu Leu Asn Asn Ile Val Lys Met Glu Glu His Ala Asp	
765 770 775 780	
tcg atc tcg gat ttc agt aga tca tca gaa tac agc aca gga agc aac	2402
Ser Ile Ser Asp Phe Ser Arg Ser Ser Glu Tyr Ser Thr Gly Ser Asn	
785 790 795	
ggt gac acg cca aga cga aag aga ata cta aaa acc aca aat gct tat	2450
Gly Asp Thr Pro Arg Arg Lys Arg Ile Leu Lys Thr Thr Asn Ala Tyr	
800 805 810	
ttc tac tgg gtc aca aga gaa caa ggc tct ttt gat tgg ttc aaa ggt	2498
Phe Tyr Trp Val Thr Arg Glu Gln Gly Ser Phe Asp Trp Phe Lys Gly	
815 820 825	
gtc atg aac gaa gtt gca gaa ctt gac caa cgg ggt gtg ata gag atg	2546
Val Met Asn Glu Val Ala Glu Leu Asp Gln Arg Gly Val Ile Glu Met	
830 835 840	
cat aac tat tta aca agt gtg tat gaa gaa ggt gat gct cgt tct gct	2594
His Asn Tyr Leu Thr Ser Val Tyr Glu Glu Gly Asp Ala Arg Ser Ala	
845 850 855 860	
ctc att aca atg gtt caa gct ctt aat cat gcc aaa aat ggt gtc gac	2642
Leu Ile Thr Met Val Gln Ala Leu Asn His Ala Lys Asn Gly Val Asp	
865 870 875	
att gtc tct ggc act agg gtc aga aca cac ttt gca aga cct aat tgg	2690
Ile Val Ser Gly Thr Arg Val Arg Thr His Phe Ala Arg Pro Asn Trp	
880 885 890	
aag aag gtt ctc aca aag cta agt tcc aag cat tgc aat gca aga aca	2738
Lys Lys Val Leu Thr Lys Leu Ser Ser Lys His Cys Asn Ala Arg Thr	
895 900 905	
gga gtg ttt tat tgc gga gta ccg gtt tta ggg aag gag ctt agc aaa	2786
Gly Val Phe Tyr Cys Gly Val Pro Val Leu Gly Lys Glu Leu Ser Lys	
910 915 920	
cta tgc aac aca ttc aat caa aaa ggt tca acc aag ttt gaa ttt cac	2834
Leu Cys Asn Thr Phe Asn Gln Lys Gly Ser Thr Lys Phe Glu Phe His	
925 930 935 940	
aag gag cat ttc taaaagacaa gaaggaagaa gccaaaagcc ctctagattc	2886
Lys Glu His Phe	
tttaatatct caaatttagc cacttatagt ataaaggcaa tctcttcact atttaattca	2946
aagtgattaa acgttaacac actgtcaaaa gtgagtgtgt taacgttttag ctccacacgt	3006
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ggtttgatag aagc

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<211> 944

<212> PRT

<213> Arabidopsis thaliana

<400> 12

Met	Lys	Pro	Phe	Ser	Lys	Asn	Asp	Arg	Arg	Arg	Trp	Ser	Phe	Asp	Ser	1	5	10	15
Val	Ser	Ala	Gly	Lys	Thr	Ala	Val	Gly	Ser	Ala	Ser	Thr	Ser	Pro	Gly	20	25	30	
Thr	Glu	Tyr	Ser	Ile	Asn	Gly	Asp	Gln	Glu	Phe	Val	Glu	Val	Thr	Ile	35	40	45	
Asp	Leu	Gln	Asp	Asp	Asp	Thr	Ile	Val	Leu	Arg	Ser	Val	Glu	Pro	Ala	50	55	60	
Thr	Ala	Ile	Asn	Val	Ile	Gly	Asp	Ile	Ser	Asp	Asp	Asn	Thr	Gly	Ile	65	70	75	80
Met	Thr	Pro	Val	Ser	Ile	Ser	Arg	Ser	Pro	Thr	Met	Lys	Arg	Thr	Ser	85	90	95	
Ser	Asn	Arg	Phe	Arg	Gln	Phe	Ser	Gln	Glu	Leu	Lys	Ala	Glu	Ala	Val	100	105	110	
Ala	Lys	Ala	Lys	Gln	Leu	Ser	Gln	Glu	Leu	Lys	Arg	Phe	Ser	Trp	Ser	115	120	125	
Arg	Ser	Phe	Ser	Gly	Asn	Leu	Thr	Thr	Thr	Ser	Thr	Ala	Ala	Asn	Gln	130	135	140	
Ser	Gly	Gly	Ala	Gly	Gly	Gly	Leu	Val	Asn	Ser	Ala	Leu	Glu	Ala	Arg	145	150	155	160
Ala	Leu	Arg	Lys	Gln	Arg	Ala	Gln	Leu	Asp	Arg	Thr	Arg	Ser	Ser	Ala	165	170	175	
Gln	Arg	Ala	Leu	Arg	Gly	Leu	Arg	Phe	Ile	Ser	Asn	Lys	Gln	Lys	Asn	180	185	190	
Val	Asp	Gly	Trp	Asn	Asp	Val	Gln	Ser	Asn	Phe	Glu	Lys	Phe	Glu	Lys	195	200	205	
Asn	Gly	Tyr	Ile	Tyr	Arg	Ser	Asp	Phe	Ala	Gln	Cys	Ile	Gly	Met	Lys	210	215	220	
Asp	Ser	Lys	Glu	Phe	Ala	Leu	Glu	Leu	Phe	Asp	Ala	Leu	Ser	Arg	Arg	225	230	235	240
Arg	Arg	Leu	Lys	Val	Glu	Lys	Ile	Asn	His	Asp	Glu	Leu	Tyr	Glu	Tyr	245	250	255	
Trp	Ser	Gln	Ile	Asn	Asp	Glu	Ser	Phe	Asp	Ser	Arg	Leu	Gln	Ile	Phe	260	265	270	
Phe	Asp	Ile	Val	Asp	Lys	Asn	Glu	Asp	Gly	Arg	Ile	Thr	Glu	Glu	Glu	275	280	285	
Val	Lys	Glu	Ile	Ile	Met	Leu	Ser	Ala	Ser	Ala	Asn	Lys	Leu	Ser	Arg	290	295	300	
Leu	Lys	Glu	Gln	Ala	Glu	Glu	Tyr	Ala	Ala	Leu	Ile	Met	Glu	Glu	Leu	305	310	315	320
Asp	Pro	Glu	Arg	Leu	Gly	Tyr	Ile	Glu	Leu	Trp	Gln	Leu	Glu	Thr	Leu	325	330	335	

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Leu	Leu	Gln	Lys	Asp	Thr	Tyr	Leu	Asn	Tyr	Ser	Gln	Ala	Leu	Ser	Tyr	340	345	350	
Thr	Ser	Gln	Ala	Leu	Ser	Gln	Asn	Leu	Gln	Gly	Leu	Arg	Gly	Lys	Ser	355	360	365	
Arg	Ile	His	Arg	Met	Ser	Ser	Asp	Phe	Val	Tyr	Ile	Met	Gln	Glu	Asn	370	375	380	
Trp	Lys	Arg	Ile	Trp	Val	Leu	Ser	Leu	Trp	Ile	Met	Ile	Met	Ile	Gly	385	390	395	400
Leu	Phe	Leu	Trp	Lys	Phe	Phe	Gln	Tyr	Lys	Gln	Lys	Asp	Ala	Phe	His	405	410	415	
Val	Met	Gly	Tyr	Cys	Leu	Leu	Thr	Ala	Lys	Gly	Ala	Ala	Glu	Thr	Leu	420	425	430	
Lys	Phe	Asn	Met	Ala	Leu	Ile	Leu	Phe	Pro	Val	Cys	Arg	Asn	Thr	Ile	435	440	445	
Thr	Trp	Leu	Arg	Ser	Thr	Arg	Leu	Ser	Tyr	Phe	Val	Pro	Phe	Asp	Asp	450	455	460	
Asn	Ile	Asn	Phe	His	Lys	Thr	Ile	Ala	Gly	Ala	Ile	Val	Val	Ala	Val	465	470	475	480
Ile	Leu	His	Ile	Gly	Asp	His	Leu	Ala	Cys	Asp	Phe	Pro	Arg	Ile	Val	485	490	495	
Arg	Ala	Thr	Glu	Tyr	Asp	Tyr	Asn	Arg	Tyr	Leu	Phe	His	Tyr	Phe	Gln	500	505	510	
Thr	Lys	Gln	Pro	Thr	Tyr	Phe	Asp	Leu	Val	Lys	Gly	Pro	Glu	Gly	Ile	515	520	525	
Thr	Gly	Ile	Leu	Met	Val	Ile	Leu	Met	Ile	Ile	Ser	Phe	Thr	Leu	Ala	530	535	540	
Thr	Arg	Trp	Phe	Arg	Arg	Asn	Leu	Val	Lys	Leu	Pro	Lys	Pro	Phe	Asp	545	550	555	560
Arg	Leu	Thr	Gly	Phe	Asn	Ala	Phe	Trp	Tyr	Ser	His	His	Leu	Phe	Val	565	570	575	
Ile	Val	Tyr	Ile	Leu	Leu	Ile	Leu	His	Gly	Ile	Phe	Leu	Tyr	Phe	Ala	580	585	590	
Lys	Pro	Trp	Tyr	Val	Arg	Thr	Thr	Trp	Met	Tyr	Leu	Ala	Val	Pro	Val	595	600	605	
Leu	Leu	Tyr	Gly	Gly	Glu	Arg	Thr	Leu	Arg	Tyr	Phe	Arg	Ser	Gly	Ser	610	615	620	
Tyr	Ser	Val	Arg	Leu	Leu	Lys	Val	Ala	Ile	Tyr	Pro	Gly	Asn	Val	Leu	625	630	635	640
Thr	Leu	Gln	Met	Ser	Lys	Pro	Thr	Gln	Phe	Arg	Tyr	Lys	Ser	Gly	Gln	645	650	655	
Tyr	Met	Phe	Val	Gln	Cys	Pro	Ala	Val	Ser	Pro	Phe	Glu	Trp	His	Pro	660	665	670	
Phe	Ser	Ile	Thr	Ser	Ala	Pro	Glu	Asp	Asp	Tyr	Ile	Ser	Ile	His	Ile	675	680	685	
Arg	Gln	Leu	Gly	Asp	Trp	Thr	Gln	Glu	Leu	Lys	Arg	Val	Phe	Ser	Glu	690	695	700	
Val	Cys	Glu	Pro	Pro	Val	Gly	Gly	Lys	Ser	Gly	Leu	Leu	Arg	Ala	Asp	705	710	715	720

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Glu Thr Thr Lys Lys Ser Leu Pro Lys Leu Leu Ile Asp Gly Pro Tyr
 725 730 735
 Gly Ala Pro Ala Gln Asp Tyr Arg Lys Tyr Asp Val Leu Leu Leu Val
 740 745 750
 Gly Leu Gly Ile Gly Ala Thr Pro Phe Ile Ser Ile Leu Lys Asp Leu
 755 760 765
 Leu Asn Asn Ile Val Lys Met Glu Glu His Ala Asp Ser Ile Ser Asp
 770 775 780
 Phe Ser Arg Ser Ser Glu Tyr Ser Thr Gly Ser Asn Gly Asp Thr Pro
 785 790 795 800
 Arg Arg Lys Arg Ile Leu Lys Thr Thr Asn Ala Tyr Phe Tyr Trp Val
 805 810 815
 Thr Arg Glu Gln Gly Ser Phe Asp Trp Phe Lys Gly Val Met Asn Glu
 820 825 830
 Val Ala Glu Leu Asp Gln Arg Gly Val Ile Glu Met His Asn Tyr Leu
 835 840 845
 Thr Ser Val Tyr Glu Glu Gly Asp Ala Arg Ser Ala Leu Ile Thr Met
 850 855 860
 Val Gln Ala Leu Asn His Ala Lys Asn Gly Val Asp Ile Val Ser Gly
 865 870 875 880
 Thr Arg Val Arg Thr His Phe Ala Arg Pro Asn Trp Lys Lys Val Leu
 885 890 895
 Thr Lys Leu Ser Ser Lys His Cys Asn Ala Arg Thr Gly Val Phe Tyr
 900 905 910
 Cys Gly Val Pro Val Leu Gly Lys Glu Leu Ser Lys Leu Cys Asn Thr
 915 920 925
 Phe Asn Gln Lys Gly Ser Thr Lys Phe Glu Phe His Lys Glu His Phe
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<212> DNA

<213> Arabidopsis thaliana

<220>

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<222> (132)..(2894)

<223> coding for NADPH oxidase

<400> 13

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 tctcgaattc g atg aaa atg aga cga ggc aat tca agt aac gac cat gaa 170
 Met Lys Met Arg Arg Gly Asn Ser Ser Asn Asp His Glu
 1 5 10
 ctt ggg att cta cga gga gct aac tcg gac acc aac tcg gac acg gag 218
 Leu Gly Ile Leu Arg Gly Ala Asn Ser Asp Thr Asn Ser Asp Thr Glu
 15 20 25
 agc atc gct agc gac cgt ggt gcc ttt agc ggt ccg ctt ggc cgg cct 266
 Ser Ile Ala Ser Asp Arg Gly Ala Phe Ser Gly Pro Leu Gly Arg Pro
 30 35 40 45

35

aaa cgt gcg tcc aag aaa aac gca aga ttc gcc gac gat ctt ccc aag	314
Lys Arg Ala Ser Lys Lys Asn Ala Arg Phe Ala Asp Asp Leu Pro Lys	
50 55 60	
aga agc aat agt gtt gct ggc ggc cgt ggt gat gac gat gag tac gtg	362
Arg Ser Asn Ser Val Ala Gly Gly Arg Gly Asp Asp Asp Glu Tyr Val	
65 70 75	
gag atc acg cta gac atc agg gac gac tcg gtg gcc gtc cat agt gtc	410
Glu Ile Thr Leu Asp Ile Arg Asp Asp Ser Val Ala Val His Ser Val	
80 85 90	
caa caa gca gct gga ggt gga ggc cac ctg gag gac ccg gag cta gcc	458
Gln Gln Ala Ala Gly Gly Gly Gly His Leu Glu Asp Pro Glu Leu Ala	
95 100 105	
ctt ctt acg aag aag act ctc gag agc agc ctc aac aac acc acc tcc	506
Leu Leu Thr Lys Lys Thr Leu Glu Ser Ser Leu Asn Asn Thr Thr Ser	
110 115 120 125	
tta tct ttc ttc cga agc acc tcc tca cgc atc aag aac gcc tcc cgc	554
Leu Ser Phe Phe Arg Ser Thr Ser Ser Arg Ile Lys Asn Ala Ser Arg	
130 135 140	
gag ctc cgc cgc gtg ttc tct aga cgt ccc tcc ccg gcc gtg cgg cgg	602
Glu Leu Arg Arg Val Phe Ser Arg Arg Pro Ser Pro Ala Val Arg Arg	
145 150 155	
ttt gac cgc acg agc tcc gcg gcc atc cac gca ctc aaa ggt ctc aag	650
Phe Asp Arg Thr Ser Ser Ala Ala Ile His Ala Leu Lys Gly Leu Lys	
160 165 170	
ttc att gcc acc aag acg gcc gca tgg ccg gcc gtc gac caa cgt ttc	698
Phe Ile Ala Thr Lys Thr Ala Ala Trp Pro Ala Val Asp Gln Arg Phe	
175 180 185	
gat aaa ctc tcc gct gat tcc aac ggc ctc tta ctc tct gcc aag ttt	746
Asp Lys Leu Ser Ala Asp Ser Asn Gly Leu Leu Leu Ser Ala Lys Phe	
190 195 200 205	
tgg gaa tgc tta gga atg aat aag gaa tct aaa gac ttc gct gac cag	794
Trp Glu Cys Leu Gly Met Asn Lys Glu Ser Lys Asp Phe Ala Asp Gln	
210 215 220	
ctc ttt aga gca tta gct cgc cgg aat aac gtc tcc ggc gat gca atc	842
Leu Phe Arg Ala Leu Ala Arg Arg Asn Asn Val Ser Gly Asp Ala Ile	
225 230 235	
aca aag gaa cag ctt agg ata ttc tgg gaa cag atc tca gac gaa agc	890
Thr Lys Glu Gln Leu Arg Ile Phe Trp Glu Gln Ile Ser Asp Glu Ser	
240 245 250	
ttt gat gcc aaa ctc caa gtc ttt ttt gac atg gtg gac aaa gat gaa	938
Phe Asp Ala Lys Leu Gln Val Phe Phe Asp Met Val Asp Lys Asp Glu	
255 260 265	
gat ggg cga gta aca gaa gaa gag gtg gct gag att att agt ctt agt	986
Asp Gly Arg Val Thr Glu Glu Glu Val Ala Glu Ile Ile Ser Leu Ser	
270 275 280 285	
gct tct gca aac aag ctc tca aat att caa aag caa gcc aaa gaa tat	1034
Ala Ser Ala Asn Lys Leu Ser Asn Ile Gln Lys Gln Ala Lys Glu Tyr	
290 295 300	
gcg gca ctg ata atg gaa gag ttg gac cca gac aat gct ggg ttt att	1082
Ala Ala Leu Ile Met Glu Glu Leu Asp Pro Asp Asn Ala Gly Phe Ile	
305 310 315	

36

atg atc gaa aac ttg gaa atg ttg cta tta caa gca ccg aac cag tcg	1130
Met Ile Glu Asn Leu Glu Met Leu Leu Leu Gln Ala Pro Asn Gln Ser	
320 325 330	
gtg cgg atg gga gac agc agg ata ctt agt cag atg tta agt cag aag	1178
Val Arg Met Gly Asp Ser Arg Ile Leu Ser Gln Met Leu Ser Gln Lys	
335 340 345	
ctt aga ccg gca aaa gag agc aac cct tta ttg aga tgg tcg gag aaa	1226
Leu Arg Pro Ala Lys Glu Ser Asn Pro Leu Leu Arg Trp Ser Glu Lys	
350 355 360 365	
atc aaa tat ttc ata ctt gat aat tgg cag aga tta tgg atc atg atg	1274
Ile Lys Tyr Phe Ile Leu Asp Asn Trp Gln Arg Leu Trp Ile Met Met	
370 375 380	
tta tgg ctt ggc atc tgt ggt ggc ctc ttt act tat aaa ttc att cag	1322
Leu Trp Leu Gly Ile Cys Gly Gly Leu Phe Thr Tyr Lys Phe Ile Gln	
385 390 395	
tac aag aac aaa gct gcc tat ggt gtg atg ggt tat tgt gtt tgt gtc	1370
Tyr Lys Asn Lys Ala Ala Tyr Gly Val Met Gly Tyr Cys Val Cys Val	
400 405 410	
gcc aaa gga ggc gcc gag act ctc aaa ttc aac atg gct ctc ata ttg	1418
Ala Lys Gly Gly Ala Glu Thr Leu Lys Phe Asn Met Ala Leu Ile Leu	
415 420 425	
ttg cct gtt tgt cga aac acc atc act tgg ctt agg aac aag acc aag	1466
Leu Pro Val Cys Arg Asn Thr Ile Thr Trp Leu Arg Asn Lys Thr Lys	
430 435 440 445	
ctt ggt act gtc gtt cct ttt gat gat agt ctt aac ttc cac aag gtt	1514
Leu Gly Thr Val Val Pro Phe Asp Asp Ser Leu Asn Phe His Lys Val	
450 455 460	
att gca agc ggg ata gtc gtc ggt gtt ttg ctc cat gcg ggt gcc cat	1562
Ile Ala Ser Gly Ile Val Val Gly Val Leu Leu His Ala Gly Ala His	
465 470 475	
tta acg tgt gat ttt cca cgt tta att gcc gcg gat gag gac acc tat	1610
Leu Thr Cys Asp Phe Pro Arg Leu Ile Ala Ala Asp Glu Asp Thr Tyr	
480 485 490	
gag ccg atg gaa aaa tac ttt ggg gat caa ccg act agc tac tgg tgg	1658
Glu Pro Met Glu Lys Tyr Phe Gly Asp Gln Pro Thr Ser Tyr Trp Trp	
495 500 505	
ttt gtg aaa gga gtg gaa gga tgg act ggc att gtg atg gtt gtg cta	1706
Phe Val Lys Gly Val Glu Gly Trp Thr Gly Ile Val Met Val Val Leu	
510 515 520 525	
atg gct ata gcc ttt aca ctc gct acg cct tgg ttc cga cgt aac aag	1754
Met Ala Ile Ala Phe Thr Leu Ala Thr Pro Trp Phe Arg Arg Asn Lys	
530 535 540	
ctt aac tta cct aac ttc ctc aag aag ctt acc ggt ttc aac gcc ttt	1802
Leu Asn Leu Pro Asn Phe Leu Lys Lys Leu Thr Gly Phe Asn Ala Phe	
545 550 555	
tgg tac acc cac cat ttg ttc atc att gtt tat gct ctt ctc att gtc	1850
Trp Tyr Thr His His Leu Phe Ile Ile Val Tyr Ala Leu Leu Ile Val	
560 565 570	
cat ggt atc aag ctc tac ctc aca aag att tgg tat cag aag acg aca	1898
His Gly Ile Lys Leu Tyr Leu Thr Lys Ile Trp Tyr Gln Lys Thr Thr	
575 580 585	

37

tgg atg tat ctt gct gta ccc atc ctt cta tat gca tct gag agg ctg	1946
Trp Met Tyr Leu Ala Val Pro Ile Leu Leu Tyr Ala Ser Glu Arg Leu	
590 595 600 605	
ctc cgt gct ttc aga tca agc atc aaa ccg gtt aag atg atc aag gtg	1994
Leu Arg Ala Phe Arg Ser Ser Ile Lys Pro Val Lys Met Ile Lys Val	
610 615 620	
gct gtt tac ccc ggg aac gtg ttg tct cta cac atg acg aag cca caa	2042
Ala Val Tyr Pro Gly Asn Val Leu Ser Leu His Met Thr Lys Pro Gln	
625 630 635	
gga ttc aaa tac aaa agt gga cag ttc atg ttg gtg aac tgc cga gcc	2090
Gly Phe Lys Tyr Lys Ser Gly Gln Phe Met Leu Val Asn Cys Arg Ala	
640 645 650	
gta tct cca ttc gaa tgg cat cct ttc tca atc aca tca gct ccc gga	2138
Val Ser Pro Phe Glu Trp His Pro Phe Ser Ile Thr Ser Ala Pro Gly	
655 660 665	
gac gat tac ctg agc gta cat atc cgc act ctc ggt gac tgg aca cgt	2186
Asp Asp Tyr Leu Ser Val His Ile Arg Thr Leu Gly Asp Trp Thr Arg	
670 675 680 685	
aag ctc agg acc gtt ttc tcc gag gtt tgc aaa cct cct acc gcc ggt	2234
Lys Leu Arg Thr Val Phe Ser Glu Val Cys Lys Pro Pro Thr Ala Gly	
690 695 700	
aaa agc ggt ctt ctc cga gca gac gga gga gat gga aac ctc ccg ttc	2282
Lys Ser Gly Leu Leu Arg Ala Asp Gly Gly Asp Gly Asn Leu Pro Phe	
705 710 715	
ccg aag gtc ctt atc gac ggt cca tac ggt gct ccc gca caa gac tac	2330
Pro Lys Val Leu Ile Asp Gly Pro Tyr Gly Ala Pro Ala Gln Asp Tyr	
720 725 730	
aag aaa tac gac gtg gta ctc ctc gta ggt ctc ggc att gga gcc acg	2378
Lys Lys Tyr Asp Val Val Leu Leu Val Gly Leu Gly Ile Gly Ala Thr	
735 740 745	
cct atg atc agt atc ctt aag gac atc atc aac aac atg aaa ggt cct	2426
Pro Met Ile Ser Ile Leu Lys Asp Ile Ile Asn Asn Met Lys Gly Pro	
750 755 760 765	
gac cgc gac agc gac att gag aac aat aac agt aac aac aat agt aaa	2474
Asp Arg Asp Ser Asp Ile Glu Asn Asn Asn Ser Asn Asn Asn Ser Lys	
770 775 780	
ggg ttt aag aca agg aaa gct tat ttc tac tgg gtg act agg gaa caa	2522
Gly Phe Lys Thr Arg Lys Ala Tyr Phe Tyr Trp Val Thr Arg Glu Gln	
785 790 795	
gga tca ttc gag tgg ttc aag gga ata atg gac gag att tcg gag tta	2570
Gly Ser Phe Glu Trp Phe Lys Gly Ile Met Asp Glu Ile Ser Glu Leu	
800 805 810	
gac gag gaa gga atc atc gag ctt cac aat tat tgc acg agt gtg tac	2618
Asp Glu Glu Gly Ile Ile Glu Leu His Asn Tyr Cys Thr Ser Val Tyr	
815 820 825	
gag gaa ggt gat gca aga gtg gct ctc att gcc atg ctt cag tcg ttg	2666
Glu Glu Gly Asp Ala Arg Val Ala Leu Ile Ala Met Leu Gln Ser Leu	
830 835 840 845	
caa cac gct aag aac ggt gtg gat gtt gtg tcg ggt aca cgt gtc aag	2714
Gln His Ala Lys Asn Gly Val Asp Val Val Ser Gly Thr Arg Val Lys	
850 855 860	

38

tcc cac ttc gct aaa cct aac tgg aga caa gtc tac aag aag atc gct 2762
 Ser His Phe Ala Lys Pro Asn Trp Arg Gln Val Tyr Lys Lys Ile Ala
 865 870 875

gtt caa cat ccc ggc aaa aga ata gga gtc ttc tac tgt gga atg cca 2810
 Val Gln His Pro Gly Lys Arg Ile Gly Val Phe Tyr Cys Gly Met Pro
 880 885 890

gga atg ata aag gaa tta aaa aat cta gct ttg gat ttt tct cga aag 2858
 Gly Met Ile Lys Glu Leu Lys Asn Leu Ala Leu Asp Phe Ser Arg Lys
 895 900 905

aca act acc aag ttt gac ttc cac aaa gag aac ttc tagattaatt 2904
 Thr Thr Thr Lys Phe Asp Phe His Lys Glu Asn Phe
 910 915 920

atatacgttg tagaaaaata aaacaagaaa caactataca aataaatatt tatttttaaatt 2964
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 aaaaaaaaaa a 3035

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<212> PRT

<213> Arabidopsis thaliana

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Leu Arg Gly Ala Asn Ser Asp Thr Asn Ser Asp Thr Glu Ser Ile Ala
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Ser Asp Arg Gly Ala Phe Ser Gly Pro Leu Gly Arg Pro Lys Arg Ala
 35 40 45

Ser Lys Lys Asn Ala Arg Phe Ala Asp Asp Leu Pro Lys Arg Ser Asn
 50 55 60

Ser Val Ala Gly Gly Arg Gly Asp Asp Asp Glu Tyr Val Glu Ile Thr
 65 70 75 80

Leu Asp Ile Arg Asp Asp Ser Val Ala Val His Ser Val Gln Gln Ala
 85 90 95

Ala Gly Gly Gly Gly His Leu Glu Asp Pro Glu Leu Ala Leu Leu Thr
 100 105 110

Lys Lys Thr Leu Glu Ser Ser Leu Asn Asn Thr Thr Ser Leu Ser Phe
 115 120 125

Phe Arg Ser Thr Ser Ser Arg Ile Lys Asn Ala Ser Arg Glu Leu Arg
 130 135 140

Arg Val Phe Ser Arg Arg Pro Ser Pro Ala Val Arg Arg Phe Asp Arg
 145 150 155 160

Thr Ser Ser Ala Ala Ile His Ala Leu Lys Gly Leu Lys Phe Ile Ala
 165 170 175

Thr Lys Thr Ala Ala Trp Pro Ala Val Asp Gln Arg Phe Asp Lys Leu
 180 185 190

Ser Ala Asp Ser Asn Gly Leu Leu Leu Ser Ala Lys Phe Trp Glu Cys
 195 200 205

Leu Gly Met Asn Lys Glu Ser Lys Asp Phe Ala Asp Gln Leu Phe Arg
 210 215 220

39

Ala	Leu	Ala	Arg	Arg	Asn	Asn	Val	Ser	Gly	Asp	Ala	Ile	Thr	Lys	Glu	225			230		235					240
Gln	Leu	Arg	Ile	Phe	Trp	Glu	Gln	Ile	Ser	Asp	Glu	Ser	Phe	Asp	Ala				245		250				255	
Lys	Leu	Gln	Val	Phe	Phe	Asp	Met	Val	Asp	Lys	Asp	Glu	Asp	Gly	Arg				260		265				270	
Val	Thr	Glu	Glu	Glu	Val	Ala	Glu	Ile	Ile	Ser	Leu	Ser	Ala	Ser	Ala				275		280				285	
Asn	Lys	Leu	Ser	Asn	Ile	Gln	Lys	Gln	Ala	Lys	Glu	Tyr	Ala	Ala	Leu				290		295				300	
Ile	Met	Glu	Glu	Leu	Asp	Pro	Asp	Asn	Ala	Gly	Phe	Ile	Met	Ile	Glu				305		310				315	
Asn	Leu	Glu	Met	Leu	Leu	Leu	Gln	Ala	Pro	Asn	Gln	Ser	Val	Arg	Met				325		330				335	
Gly	Asp	Ser	Arg	Ile	Leu	Ser	Gln	Met	Leu	Ser	Gln	Lys	Leu	Arg	Pro				340		345				350	
Ala	Lys	Glu	Ser	Asn	Pro	Leu	Leu	Arg	Trp	Ser	Glu	Lys	Ile	Lys	Tyr				355		360				365	
Phe	Ile	Leu	Asp	Asn	Trp	Gln	Arg	Leu	Trp	Ile	Met	Met	Leu	Trp	Leu				370		375				380	
Gly	Ile	Cys	Gly	Gly	Leu	Phe	Thr	Tyr	Lys	Phe	Ile	Gln	Tyr	Lys	Asn				385		390				395	
Lys	Ala	Ala	Tyr	Gly	Val	Met	Gly	Tyr	Cys	Val	Cys	Val	Ala	Lys	Gly				405		410				415	
Gly	Ala	Glu	Thr	Leu	Lys	Phe	Asn	Met	Ala	Leu	Ile	Leu	Leu	Pro	Val				420		425				430	
Cys	Arg	Asn	Thr	Ile	Thr	Trp	Leu	Arg	Asn	Lys	Thr	Lys	Leu	Gly	Thr				435		440				445	
Val	Val	Pro	Phe	Asp	Asp	Ser	Leu	Asn	Phe	His	Lys	Val	Ile	Ala	Ser				450		455				460	
Gly	Ile	Val	Val	Gly	Val	Leu	Leu	His	Ala	Gly	Ala	His	Leu	Thr	Cys				465		470				475	
Asp	Phe	Pro	Arg	Leu	Ile	Ala	Ala	Asp	Glu	Asp	Thr	Tyr	Glu	Pro	Met				485		490				495	
Glu	Lys	Tyr	Phe	Gly	Asp	Gln	Pro	Thr	Ser	Tyr	Trp	Trp	Phe	Val	Lys				500		505				510	
Gly	Val	Glu	Gly	Trp	Thr	Gly	Ile	Val	Met	Val	Val	Leu	Met	Ala	Ile				515		520				525	
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His	His	Leu	Phe	Ile	Ile	Val	Tyr	Ala	Leu	Leu	Ile	Val	His	Gly	Ile				565		570				575	
Lys	Leu	Tyr	Leu	Thr	Lys	Ile	Trp	Tyr	Gln	Lys	Thr	Thr	Trp	Met	Tyr				580		585				590	
Leu	Ala	Val	Pro	Ile	Leu	Leu	Tyr	Ala	Ser	Glu	Arg	Leu	Leu	Arg	Ala				595		600				605	

40

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Pro	Gly	Asn	Val	Leu	Ser	Leu	His	Met	Thr	Lys	Pro	Gln	Gly	Phe	Lys
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Tyr	Lys	Ser	Gly	Gln	Phe	Met	Leu	Val	Asn	Cys	Arg	Ala	Val	Ser	Pro
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Ser	Ile	Leu	Lys	Asp	Ile	Ile	Asn	Asn	Met	Lys	Gly	Pro	Asp	Arg	Asp
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Ala	Lys	Pro	Asn	Trp	Arg	Gln	Val	Tyr	Lys	Lys	Ile	Ala	Val	Gln	His
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Pro	Gly	Lys	Arg	Ile	Gly	Val	Phe	Tyr	Cys	Gly	Met	Pro	Gly	Met	Ile
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Lys	Glu	Leu	Lys	Asn	Leu	Ala	Leu	Asp	Phe	Ser	Arg	Lys	Thr	Thr	Thr
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Lys	Phe	Asp	Phe	His	Lys	Glu	Asn	Phe							
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<212> DNA

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<220>

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<222> (313)..(3129)

<223> coding for NADPH oxidase

41

<400> 15

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ataccaatta ttaatctttt gtcactctttt tccttctcgc gttcatatat actggaatat 180
acatctttttt ttcaacctat cttcttttcat tttcaagaat tcggggttcca taaatagtag 240
gttcactact tttatttcaa cctccttaaa gtttattcat tcatattttt tctcaaagaa 300
aaaactatag aa atg caa aat tcg gaa aat cat cat ccg cac cac cag cac 351
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cac cat tcg gac aca gag ata att gga aat gat aga gcg tcg tac agt 399
His His Ser Asp Thr Glu Ile Ile Gly Asn Asp Arg Ala Ser Tyr Ser
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Gly Pro Leu Ser Gly Pro Leu Asn Lys Arg Gly Gly Lys Lys Ser Ala
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aga ttt aac att cct gaa tct acc gac atc gga acc agt gtc gga acc 495
Arg Phe Asn Ile Pro Glu Ser Thr Asp Ile Gly Thr Ser Val Gly Thr
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ggc ggc aag tcc aat gat gat gcg tac gtt gaa atc act ctc gat gtc 543
Gly Gly Lys Ser Asn Asp Asp Ala Tyr Val Glu Ile Thr Leu Asp Val
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cgc gaa gat tcc gtc gct gtc cac agt gtc aaa act gcc ggc ggt gat 591
Arg Glu Asp Ser Val Ala Val His Ser Val Lys Thr Ala Gly Gly Asp
              80              85              90
gac gtg gaa gat ccc gag ctg gct tta ttg gct aaa ggc tta gag aag 639
Asp Val Glu Asp Pro Glu Leu Ala Leu Leu Ala Lys Gly Leu Glu Lys
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aag tcc act tta gga tct tca ctt gtt cga aat gct tcg tct aga att 687
Lys Ser Thr Leu Gly Ser Ser Leu Val Arg Asn Ala Ser Ser Arg Ile
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cca att cct act gga agg ttc gac agg aat aaa tca gct gct gct cat 783
Pro Ile Pro Thr Gly Arg Phe Asp Arg Asn Lys Ser Ala Ala Ala His
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gct ctt aaa ggt ctc aag ttt att agt aag acc gac ggc ggc gct ggt 831
Ala Leu Lys Gly Leu Lys Phe Ile Ser Lys Thr Asp Gly Gly Ala Gly
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Trp Ala Ala Val Glu Lys Arg Phe Asp Glu Ile Thr Ala Ser Thr Thr
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gag tct aag gaa ttt gct gtt gag cta tat gat gcg cta gct cgg agg 975
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aga aac att aca act gat tcc att aac aaa gca cag ctc aaa gag ttc 1023
Arg Asn Ile Thr Thr Asp Ser Ile Asn Lys Ala Gln Leu Lys Glu Phe
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42

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240 245 250	
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Phe Asp Met Val Asp Lys Asp Ala Asp Gly Arg Ile Thr Glu Glu Glu	
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gtc aga gag att ata ggc ctt agc gcg tcg gcc aac agg ctg tca aca	1167
Val Arg Glu Ile Ile Gly Leu Ser Ala Ser Ala Asn Arg Leu Ser Thr	
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Ile Gln Lys Gln Ala Asp Glu Tyr Ala Ala Met Ile Met Glu Glu Leu	
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350 355 360 365	
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Asn Trp Gln Arg Val Trp Val Leu Leu Leu Trp Ile Gly Ile Met Ala	
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Gly Leu Phe Thr Trp Lys Tyr Ile Gln Tyr Lys Glu Lys Ala Ala Tyr	
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Lys Val Met Gly Pro Cys Val Cys Phe Ala Lys Gly Ala Ala Glu Thr	
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Leu Lys Leu Asn Met Ala Ile Ile Leu Phe Pro Val Cys Arg Asn Thr	
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430 435 440 445	
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Gly Val Gly Ile His Gly Leu Ser His Leu Thr Cys Asp Phe Pro Arg	
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Leu Leu Asn Ala Ser Glu Glu Glu Tyr Glu Pro Met Lys Tyr Tyr Phe	
480 485 490	
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Gly Asp Gln Pro Glu Ser Tyr Trp Trp Phe Ile Lys Gly Val Glu Gly	
495 500 505	

43

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His	Lys	Leu	Thr	Gly	Xaa	Asn	Ala	Phe	Trp	Tyr	Ser	His	His	Leu	Phe	
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cca	atc	ata	ctc	tat	gct	agt	gaa	agg	ttg	att	agg	gca	ttc	agg	tca	2127
Pro	Ile	Ile	Leu	Tyr	Ala	Ser	Glu	Arg	Leu	Ile	Arg	Ala	Phe	Arg	Ser	
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Ser	Ile	Lys	Ala	Val	Lys	Ile	Leu	Lys	Val	Ala	Val	Tyr	Pro	Gly	Asn	
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cat	cca	ttt	tca	att	act	tcg	gcc	cca	gga	gat	gac	tat	ctc	agt	gtc	2319
His	Pro	Phe	Ser	Ile	Thr	Ser	Ala	Pro	Gly	Asp	Asp	Tyr	Leu	Ser	Val	
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cat	att	cga	act	ctt	ggt	gat	tgg	acc	agg	caa	ctt	aaa	act	gtt	ttc	2367
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tcc	gag	gtt	tgc	cag	cca	cca	cct	aat	gga	aaa	agt	gga	ctc	ctc	aga	2415
Ser	Glu	Val	Cys	Gln	Pro	Pro	Pro	Asn	Gly	Lys	Ser	Gly	Leu	Leu	Arg	
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gct	gac	tac	ttg	caa	gga	gag	aat	aat	cct	aat	ttc	cca	agg	gtg	tta	2463
Ala	Asp	Tyr	Leu	Gln	Gly	Glu	Asn	Asn	Pro	Asn	Phe	Pro	Arg	Val	Leu	
			705					710					715			
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Ile	Asp	Gly	Pro	Tyr	Gly	Ala	Pro	Ala	Gln	Asp	Tyr	Lys	Lys	Tyr	Glu	
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Val	Val	Leu	Leu	Val	Gly	Leu	Gly	Ile	Gly	Ala	Thr	Pro	Met	Ile	Ser	
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Ile	Val	Lys	Asp	Ile	Val	Asn	Asn	Met	Lys	Ala	Met	Asp	Glu	Glu	Glu	
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aat	tcc	ttg	gaa	gat	gga	cac	aat	aat	aat	atg	gca	cca	aat	tct	agc	2655
Asn	Ser	Leu	Glu	Asp	Gly	His	Asn	Asn	Asn	Met	Ala	Pro	Asn	Ser	Ser	
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44

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 850 855 860
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 35 40 45
 Ile Pro Glu Ser Thr Asp Ile Gly Thr Ser Val Gly Thr Gly Gly Lys
 50 55 60
 Ser Asn Asp Asp Ala Tyr Val Glu Ile Thr Leu Asp Val Arg Glu Asp
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45

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Leu	Gly	Ser	Ser	Leu	Val	Arg	Asn	Ala	Ser	Ser	Arg	Ile	Arg	Gln	Val	
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			180					185					190			
Pro	Arg	Ala	Lys	Phe	Gly	Glu	Cys	Ile	Gly	Met	Asn	Lys	Glu	Ser	Lys	
		195					200					205				
Glu	Phe	Ala	Val	Glu	Leu	Tyr	Asp	Ala	Leu	Ala	Arg	Arg	Arg	Asn	Ile	
	210					215					220					
Thr	Thr	Asp	Ser	Ile	Asn	Lys	Ala	Gln	Leu	Lys	Glu	Phe	Trp	Asp	Gln	
225					230					235					240	
Val	Ala	Asp	Gln	Ser	Phe	Asp	Ser	Arg	Leu	Gln	Thr	Phe	Phe	Asp	Met	
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Gln	Ala	Asp	Glu	Tyr	Ala	Ala	Met	Ile	Met	Glu	Glu	Leu	Asp	Pro	Asn	
	290					295					300					
Asn	Leu	Gly	Tyr	Ile	Met	Ile	Glu	Asn	Leu	Glu	Met	Leu	Leu	Leu	Gln	
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Ala	Pro	Asn	Gln	Ser	Val	Gln	Arg	Gly	Gly	Glu	Ser	Arg	Asn	Leu	Ser	
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Gln	Met	Leu	Ser	Gln	Lys	Leu	Lys	His	Thr	Gln	Glu	Arg	Asn	Pro	Ile	
			340					345					350			
Val	Arg	Trp	Tyr	Lys	Ser	Phe	Met	Tyr	Phe	Leu	Leu	Asp	Asn	Trp	Gln	
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						375					380					
Thr	Trp	Lys	Tyr	Ile	Gln	Tyr	Lys	Glu	Lys	Ala	Ala	Tyr	Lys	Val	Met	
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Ile	His	Gly	Leu	Ser	His	Leu	Thr	Cys	Asp	Phe	Pro	Arg	Leu	Leu	Asn	465	470	475			480
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Pro	Glu	Ser	Tyr	Trp	Trp	Phe	Ile	Lys	Gly	Val	Glu	Gly	Val	Thr	Gly		500	505			510
Ile	Ile	Met	Val	Val	Leu	Met	Ala	Ile	Ala	Phe	Thr	Leu	Ala	Thr	Pro		515	520			525
Trp	Phe	Arg	Arg	Asn	Arg	Val	Ser	Leu	Pro	Lys	Pro	Phe	His	Lys	Leu		530	535			540
Thr	Gly	Xaa	Asn	Ala	Phe	Trp	Tyr	Ser	His	His	Leu	Phe	Val	Ile	Val		545	550			555
Tyr	Thr	Leu	Phe	Ile	Val	His	Gly	Glu	Lys	Leu	Tyr	Ile	Thr	Lys	Asp		565	570			575
Trp	Tyr	Lys	Arg	Thr	Asp	Met	Asp	Val	Leu	Leu	Thr	Ile	Pro	Ile	Ile		580	585			590
Leu	Tyr	Ala	Ser	Glu	Arg	Leu	Ile	Arg	Ala	Phe	Arg	Ser	Ser	Ile	Lys		595	600			605
Ala	Val	Lys	Ile	Leu	Lys	Val	Ala	Val	Tyr	Pro	Gly	Asn	Val	Leu	Ala		610	615			620
Leu	His	Met	Ser	Lys	Pro	Gln	Gly	Tyr	Lys	Tyr	Lys	Ser	Gly	Gln	Tyr		625	630			635
Met	Phe	Val	Asn	Cys	Ala	Ala	Val	Ser	Pro	Phe	Glu	Trp	His	Pro	Phe		645	650			655
Ser	Ile	Thr	Ser	Ala	Pro	Gly	Asp	Asp	Tyr	Leu	Ser	Val	His	Ile	Arg		660	665			670
Thr	Leu	Gly	Asp	Trp	Thr	Arg	Gln	Leu	Lys	Thr	Val	Phe	Ser	Glu	Val		675	680			685
Cys	Gln	Pro	Pro	Pro	Asn	Gly	Lys	Ser	Gly	Leu	Leu	Arg	Ala	Asp	Tyr		690	695			700
Leu	Gln	Gly	Glu	Asn	Asn	Pro	Asn	Phe	Pro	Arg	Val	Leu	Ile	Asp	Gly		705	710			715
Pro	Tyr	Gly	Ala	Pro	Ala	Gln	Asp	Tyr	Lys	Lys	Tyr	Glu	Val	Val	Leu		725	730			735
Leu	Val	Gly	Leu	Gly	Ile	Gly	Ala	Thr	Pro	Met	Ile	Ser	Ile	Val	Lys		740	745			750
Asp	Ile	Val	Asn	Asn	Met	Lys	Ala	Met	Asp	Glu	Glu	Glu	Asn	Ser	Leu		755	760			765
Glu	Asp	Gly	His	Asn	Asn	Asn	Met	Ala	Pro	Asn	Ser	Ser	Pro	Asn	Ile		770	775			780
Ala	Lys	Asn	Lys	Gly	Asn	Lys	Ser	Gly	Ser	Ala	Ser	Gly	Gly	Asn	Asn		785	790			795
Phe	Asn	Thr	Arg	Arg	Ala	Tyr	Phe	Tyr	Trp	Val	Thr	Arg	Glu	Gln	Gly		805	810			815
Ser	Phe	Asp	Trp	Phe	Lys	Gly	Ile	Met	Asn	Glu	Ala	Ala	Glu	Met	Asp		820	825			830
His	Lys	Gly	Val	Ile	Glu	Met	His	Asn	Tyr	Cys	Thr	Ser	Val	Tyr	Glu		835	840			845

47

Glu	Gly	Asp	Ala	Arg	Ser	Ala	Leu	Ile	Thr	Met	Leu	Gln	Ser	Leu	His
850						855					860				
His	Ala	Lys	Asn	Gly	Val	Asp	Ile	Val	Ser	Gly	Thr	Arg	Val	Lys	Ser
865					870					875					880
His	Phe	Ala	Lys	Pro	Asn	Trp	Arg	Asn	Val	Tyr	Lys	Arg	Ile	Ala	Leu
				885					890					895	
Asn	His	Pro	Glu	Ala	Lys	Val	Gly	Val	Phe	Tyr	Cys	Gly	Ala	Pro	Ala
			900					905					910		
Leu	Thr	Lys	Glu	Leu	Arg	Gln	His	Ala	Leu	Asp	Phe	Ser	His	Lys	Thr
		915					920					925			
Ser	Thr	Lys	Phe	Asp	Phe	His	Lys	Glu	Asn	Phe					
		930				935									

<210> 17

<211> 2532

<212> DNA

<213> Oryza sativa

<220>

<221> CDS

<222> (1)..(2529)

<223> coding for NADPH oxidase

<400> 17

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Met	Ala	Ser	Pro	Tyr	Asp	His	Gln	Ser	Pro	His	Ala	Gln	His	Pro	Ser	
1				5				10						15		
ggg	ttg	ccg	agg	ccg	ccg	ggg	gcg	ggg	gcg	ggt	gcg	gcg	gcg	ggc	ggg	96
Gly	Leu	Pro	Arg	Pro	Pro	Gly	Ala	Gly	Ala	Gly	Ala	Ala	Ala	Gly	Gly	
			20				25						30			
ttc	gcg	cgg	ggg	ctg	atg	aag	cag	ccg	tcg	cgg	ctg	gcg	tcc	ggg	gtg	144
Phe	Ala	Arg	Gly	Leu	Met	Lys	Gln	Pro	Ser	Arg	Leu	Ala	Ser	Gly	Val	
		35				40					45					
agg	cag	ttc	gcg	tcg	agg	gtg	tcg	atg	aag	gtg	ccg	gag	ggg	gtg	ggg	192
Arg	Gln	Phe	Ala	Ser	Arg	Val	Ser	Met	Lys	Val	Pro	Glu	Gly	Val	Gly	
	50				55					60						
ggg	atg	cgg	ccc	ggt	ggc	ggg	agg	atg	acg	cgg	atg	cag	tcc	agc	gcg	240
Gly	Met	Arg	Pro	Gly	Gly	Gly	Arg	Met	Thr	Arg	Met	Gln	Ser	Ser	Ala	
65				70				75							80	
cag	gtg	ggg	ctc	cgg	ggg	ctc	cgc	ttc	ctc	gac	aag	acg	tcc	ggc	ggg	288
Gln	Val	Gly	Leu	Arg	Gly	Leu	Arg	Phe	Leu	Asp	Lys	Thr	Ser	Gly	Gly	
			85				90						95			
aag	gag	ggg	tgg	aag	tcc	gtc	gag	cgc	cgc	ttc	gac	gag	atg	aac	cgc	336
Lys	Glu	Gly	Trp	Lys	Ser	Val	Glu	Arg	Arg	Phe	Asp	Glu	Met	Asn	Arg	
		100				105						110				
aac	ggc	cgc	ctc	ccc	aag	gag	agc	ttc	ggc	aag	tgc	atc	ggc	atg	ggg	384
Asn	Gly	Arg	Leu	Pro	Lys	Glu	Ser	Phe	Gly	Lys	Cys	Ile	Gly	Met	Gly	
		115				120						125				
gac	tcc	aag	gag	ttc	gcc	ggc	gag	ctg	ttc	gtg	gcg	ctg	gcg	cgg	cgg	432
Asp	Ser	Lys	Glu	Phe	Ala	Gly	Glu	Leu	Phe	Val	Ala	Leu	Ala	Arg	Arg	
		130				135					140					
agg	aac	ctg	gag	ccg	gag	gac	ggc	atc	acc	aag	gag	cag	ctc	aag	gag	480
Arg	Asn	Leu	Glu	Pro	Glu	Asp	Gly	Ile	Thr	Lys	Glu	Gln	Leu	Lys	Glu	
145					150					155					160	

48

ttc	tgg	gag	gag	atg	acc	gac	cag	aac	ttc	gac	tcg	cgg	ctt	cgc	att	528
Phe	Trp	Glu	Glu	Met	Thr	Asp	Gln	Asn	Phe	Asp	Ser	Arg	Leu	Arg	Ile	
				165					170						175	
ttc	ttt	gac	atg	tgc	gac	aag	aat	ggc	gat	ggg	atg	ctc	acg	gaa	gat	576
Phe	Phe	Asp	Met	Cys	Asp	Lys	Asn	Gly	Asp	Gly	Met	Leu	Thr	Glu	Asp	
			180					185						190		
gag	gtc	aag	gag	gtt	att	ata	ctg	agt	gcg	tcg	gcg	aac	aag	ctg	gcg	624
Glu	Val	Lys	Glu	Val	Ile	Ile	Leu	Ser	Ala	Ser	Ala	Asn	Lys	Leu	Ala	
			195				200					205				
aag	ctg	aag	gga	cac	gcg	gcg	acg	tac	gcg	tcg	ctg	atc	atg	gag	gag	672
Lys	Leu	Lys	Gly	His	Ala	Ala	Thr	Tyr	Ala	Ser	Leu	Ile	Met	Glu	Glu	
	210					215					220					
ctg	gac	ccg	gac	gac	cgc	ggg	tac	atc	gag	atc	tgg	cag	ctg	gag	acg	720
Leu	Asp	Pro	Asp	Asp	Arg	Gly	Tyr	Ile	Glu	Ile	Trp	Gln	Leu	Glu	Thr	
225					230					235					240	
ctg	ctg	cgc	ggc	atg	gtg	agc	gcg	cag	gcg	gcg	ccg	gag	aag	atg	aag	768
Leu	Leu	Arg	Gly	Met	Val	Ser	Ala	Gln	Ala	Ala	Pro	Glu	Lys	Met	Lys	
				245				250						255		
cgg	acg	acg	tcg	agc	ctc	gcg	agg	acg	atg	atc	ccg	tcg	cgg	tac	cgg	816
Arg	Thr	Thr	Ser	Ser	Leu	Ala	Arg	Thr	Met	Ile	Pro	Ser	Arg	Tyr	Arg	
			260					265					270			
agc	ccg	ctg	aag	cgg	cac	gtg	tcc	agg	acg	gtg	gac	ttc	gtg	cac	gag	864
Ser	Pro	Leu	Lys	Arg	His	Val	Ser	Arg	Thr	Val	Asp	Phe	Val	His	Glu	
		275					280					285				
aac	tgg	aag	cgg	atc	tgg	ctc	gtc	gcg	ctg	tgg	ctc	gcc	gtc	aac	gtc	912
Asn	Trp	Lys	Arg	Ile	Trp	Leu	Val	Ala	Leu	Trp	Leu	Ala	Val	Asn	Val	
	290					295					300					
ggc	ctc	ttc	gcc	tac	aag	ttc	gag	cag	tac	gag	cgg	cgc	gcc	gcg	ttc	960
Gly	Leu	Phe	Ala	Tyr	Lys	Phe	Glu	Gln	Tyr	Glu	Arg	Arg	Ala	Ala	Phe	
305					310					315					320	
cag	gtg	atg	ggc	cac	tgc	gtg	tgc	gtg	gcc	aag	ggc	gcc	gcc	gag	gtg	1008
Gln	Val	Met	Gly	His	Cys	Val	Cys	Val	Ala	Lys	Gly	Ala	Ala	Glu	Val	
				325				330						335		
ctc	aag	ctc	aac	atg	gcg	ctc	atc	ctc	ctc	ccc	gtg	tgc	cgg	aac	acg	1056
Leu	Lys	Leu	Asn	Met	Ala	Leu	Ile	Leu	Leu	Pro	Val	Cys	Arg	Asn	Thr	
			340					345					350			
ctc	acc	acg	ctc	agg	tcc	acg	gcg	ctc	agc	cac	gtc	atc	ccc	ttc	gac	1104
Leu	Thr	Thr	Leu	Arg	Ser	Thr	Ala	Leu	Ser	His	Val	Ile	Pro	Phe	Asp	
			355				360					365				
gac	aac	atc	aac	ttc	cac	aag	gtg	atc	gcg	gcg	acc	atc	gcc	gcc	gcc	1152
Asp	Asn	Ile	Asn	Phe	His	Lys	Val	Ile	Ala	Ala	Thr	Ile	Ala	Ala	Ala	
	370					375					380					
acc	gcc	gtc	cac	acg	ctg	gcg	cac	gtc	acc	tgc	gac	ttc	ccg	agg	ctg	1200
Thr	Ala	Val	His	Thr	Leu	Ala	His	Val	Thr	Cys	Asp	Phe	Pro	Arg	Leu	
385					390					395					400	
atc	aac	tgc	ccc	agc	gac	aag	ttc	atg	gcg	acg	ctg	ggg	ccg	aac	ttc	1248
Ile	Asn	Cys	Pro	Ser	Asp	Lys	Phe	Met	Ala	Thr	Leu	Gly	Pro	Asn	Phe	
				405				410						415		
ggg	tac	agg	cag	ccg	acg	tac	gcc	gac	ctg	ctg	gag	agc	gcc	ccc	ggc	1296
Gly	Tyr	Arg	Gln	Pro	Thr	Tyr	Ala	Asp	Leu	Leu	Glu	Ser	Ala	Pro	Gly	
			420					425					430			

49

gtc acc ggc atc ctc atg atc atc atc atg tcc ttc tcc ttc acg ctg	1344
Val Thr Gly Ile Leu Met Ile Ile Ile Met Ser Phe Ser Phe Thr Leu	
435 440 445	
gcc acg cac tcc ttc cgc cgg agc gtc gtc aag ctg ccg tcg ccg ctg	1392
Ala Thr His Ser Phe Arg Arg Ser Val Val Lys Leu Pro Ser Pro Leu	
450 455 460	
cac cac ctc gcc ggc ttc aac gcc ttc tgg tac gcg cac cac ctc ctg	1440
His His Leu Ala Gly Phe Asn Ala Phe Trp Tyr Ala His His Leu Leu	
465 470 475 480	
gtg ctc gcc tac gtc ctc ctc gtc gtg cac tcc tac ttc ata ttc ctc	1488
Val Leu Ala Tyr Val Leu Leu Val Val His Ser Tyr Phe Ile Phe Leu	
485 490 495	
acc agg gag tgg tac aag aaa acg aca tgg atg tac ctg ata gtc cca	1536
Thr Arg Glu Trp Tyr Lys Lys Thr Thr Trp Met Tyr Leu Ile Val Pro	
500 505 510	
gtg ctc ttc tat gca tgc gag aga acg atc aga aaa gtt cga gag aac	1584
Val Leu Phe Tyr Ala Cys Glu Arg Thr Ile Arg Lys Val Arg Glu Asn	
515 520 525	
aac tac cgc gtg agc atc gtc aag gca gcg att tac cca gga aat gtg	1632
Asn Tyr Arg Val Ser Ile Val Lys Ala Ala Ile Tyr Pro Gly Asn Val	
530 535 540	
ctc tct ctt cac atg aag aag ccg ccg ggt ttc aag tac aag agc ggg	1680
Leu Ser Leu His Met Lys Lys Pro Pro Gly Phe Lys Tyr Lys Ser Gly	
545 550 555 560	
atg tac ctg ttt gtg aag tgc cct gat gtc tct cct ttc gaa tgg cat	1728
Met Tyr Leu Phe Val Lys Cys Pro Asp Val Ser Pro Phe Glu Trp His	
565 570 575	
ccc ttc tcc atc act tct gca cct gga gat gac tac ctg agt gtg cat	1776
Pro Phe Ser Ile Thr Ser Ala Pro Gly Asp Asp Tyr Leu Ser Val His	
580 585 590	
atc cgt aca cta ggt gac tgg acg act gaa ctc aga aac ctg ttt ggg	1824
Ile Arg Thr Leu Gly Asp Trp Thr Thr Glu Leu Arg Asn Leu Phe Gly	
595 600 605	
aag gct tgc gag gca cag gtt act tct aag aag gct acc ctt tca aga	1872
Lys Ala Cys Glu Ala Gln Val Thr Ser Lys Lys Ala Thr Leu Ser Arg	
610 615 620	
ctt gaa act aca gtt gtg gcg gac gct cag aca gag gat act agg ttt	1920
Leu Glu Thr Thr Val Val Ala Asp Ala Gln Thr Glu Asp Thr Arg Phe	
625 630 635 640	
cct aag gtc ctt att gat ggg ccc tat ggt gca ccg gcg caa aac tac	1968
Pro Lys Val Leu Ile Asp Gly Pro Tyr Gly Ala Pro Ala Gln Asn Tyr	
645 650 655	
aag aag tat gac att ctt ttg ctt att ggt ctt gga att ggt gct act	2016
Lys Lys Tyr Asp Ile Leu Leu Leu Ile Gly Leu Gly Ile Gly Ala Thr	
660 665 670	
cct ttc atc agc att ctg aag gat ctg ttg aac aac att aaa tcc aac	2064
Pro Phe Ile Ser Ile Leu Lys Asp Leu Leu Asn Asn Ile Lys Ser Asn	
675 680 685	
gaa gag gtg gaa agc ata cat ggt tct gag ata ggc agc ttc aag aac	2112
Glu Glu Val Glu Ser Ile His Gly Ser Glu Ile Gly Ser Phe Lys Asn	
690 695 700	

50

aat ggg cca gga aga gct tac ttc tac tgg gtg acc aga gag caa ggg 2160
 Asn Gly Pro Gly Arg Ala Tyr Phe Tyr Trp Val Thr Arg Glu Gln Gly
 705 710 715 720
 tcc ttc gag tgg ttt aaa gga gtc atg aac gat gtc gct gaa agt gat 2208
 Ser Phe Glu Trp Phe Lys Gly Val Met Asn Asp Val Ala Glu Ser Asp
 725 730 735
 cac aat aat att ata gag atg cac aat tac ctg acc agc gtg tat gaa 2256
 His Asn Asn Ile Ile Glu Met His Asn Tyr Leu Thr Ser Val Tyr Glu
 740 745 750
 gaa ggc gac gca agg tca gct ttg att gcc atg gtt cag tca ctt caa 2304
 Glu Gly Asp Ala Arg Ser Ala Leu Ile Ala Met Val Gln Ser Leu Gln
 755 760 765
 cat gcc aaa aat ggt gtg gat atc gtc tcc ggc agc agg att cgc aca 2352
 His Ala Lys Asn Gly Val Asp Ile Val Ser Gly Ser Arg Ile Arg Thr
 770 775 780
 cat ttt gcg agg cct aac tgg aga aag gtg ttc tct gac ttg gcg aat 2400
 His Phe Ala Arg Pro Asn Trp Arg Lys Val Phe Ser Asp Leu Ala Asn
 785 790 795 800
 gcc cac aaa aac tca cgc ata ggt gtt ttc tat tgt gga tcc cct aca 2448
 Ala His Lys Asn Ser Arg Ile Gly Val Phe Tyr Cys Gly Ser Pro Thr
 805 810 815
 ctc acg aaa caa ctc aag gat ctt tca aaa gaa ttc agc cag aca acc 2496
 Leu Thr Lys Gln Leu Lys Asp Leu Ser Lys Glu Phe Ser Gln Thr Thr
 820 825 830
 aca act aga ttc cac ttc cac aag gaa aac ttt taa 2532
 Thr Thr Arg Phe His Phe His Lys Glu Asn Phe
 835 840

<210> 18

<211> 843

<212> PRT

<213> Oryza sativa

<400> 18

Met Ala Ser Pro Tyr Asp His Gln Ser Pro His Ala Gln His Pro Ser
 1 5 10 15
 Gly Leu Pro Arg Pro Pro Gly Ala Gly Ala Gly Ala Ala Ala Gly Gly
 20 25 30
 Phe Ala Arg Gly Leu Met Lys Gln Pro Ser Arg Leu Ala Ser Gly Val
 35 40 45
 Arg Gln Phe Ala Ser Arg Val Ser Met Lys Val Pro Glu Gly Val Gly
 50 55 60
 Gly Met Arg Pro Gly Gly Gly Arg Met Thr Arg Met Gln Ser Ser Ala
 65 70 75 80
 Gln Val Gly Leu Arg Gly Leu Arg Phe Leu Asp Lys Thr Ser Gly Gly
 85 90 95
 Lys Glu Gly Trp Lys Ser Val Glu Arg Arg Phe Asp Glu Met Asn Arg
 100 105 110
 Asn Gly Arg Leu Pro Lys Glu Ser Phe Gly Lys Cys Ile Gly Met Gly
 115 120 125
 Asp Ser Lys Glu Phe Ala Gly Glu Leu Phe Val Ala Leu Ala Arg Arg
 130 135 140

51

Arg	Asn	Leu	Glu	Pro	Glu	Asp	Gly	Ile	Thr	Lys	Glu	Gln	Leu	Lys	Glu	145	150	155	160
Phe	Trp	Glu	Glu	Met	Thr	Asp	Gln	Asn	Phe	Asp	Ser	Arg	Leu	Arg	Ile	165	170	175	
Phe	Phe	Asp	Met	Cys	Asp	Lys	Asn	Gly	Asp	Gly	Met	Leu	Thr	Glu	Asp	180	185	190	
Glu	Val	Lys	Glu	Val	Ile	Ile	Leu	Ser	Ala	Ser	Ala	Asn	Lys	Leu	Ala	195	200	205	
Lys	Leu	Lys	Gly	His	Ala	Ala	Thr	Tyr	Ala	Ser	Leu	Ile	Met	Glu	Glu	210	215	220	
Leu	Asp	Pro	Asp	Asp	Arg	Gly	Tyr	Ile	Glu	Ile	Trp	Gln	Leu	Glu	Thr	225	230	235	240
Leu	Leu	Arg	Gly	Met	Val	Ser	Ala	Gln	Ala	Ala	Pro	Glu	Lys	Met	Lys	245	250	255	
Arg	Thr	Thr	Ser	Ser	Leu	Ala	Arg	Thr	Met	Ile	Pro	Ser	Arg	Tyr	Arg	260	265	270	
Ser	Pro	Leu	Lys	Arg	His	Val	Ser	Arg	Thr	Val	Asp	Phe	Val	His	Glu	275	280	285	
Asn	Trp	Lys	Arg	Ile	Trp	Leu	Val	Ala	Leu	Trp	Leu	Ala	Val	Asn	Val	290	295	300	
Gly	Leu	Phe	Ala	Tyr	Lys	Phe	Glu	Gln	Tyr	Glu	Arg	Arg	Ala	Ala	Phe	305	310	315	320
Gln	Val	Met	Gly	His	Cys	Val	Cys	Val	Ala	Lys	Gly	Ala	Ala	Glu	Val	325	330	335	
Leu	Lys	Leu	Asn	Met	Ala	Leu	Ile	Leu	Leu	Pro	Val	Cys	Arg	Asn	Thr	340	345	350	
Leu	Thr	Thr	Leu	Arg	Ser	Thr	Ala	Leu	Ser	His	Val	Ile	Pro	Phe	Asp	355	360	365	
Asp	Asn	Ile	Asn	Phe	His	Lys	Val	Ile	Ala	Ala	Thr	Ile	Ala	Ala	Ala	370	375	380	
Thr	Ala	Val	His	Thr	Leu	Ala	His	Val	Thr	Cys	Asp	Phe	Pro	Arg	Leu	385	390	395	400
Ile	Asn	Cys	Pro	Ser	Asp	Lys	Phe	Met	Ala	Thr	Leu	Gly	Pro	Asn	Phe	405	410	415	
Gly	Tyr	Arg	Gln	Pro	Thr	Tyr	Ala	Asp	Leu	Leu	Glu	Ser	Ala	Pro	Gly	420	425	430	
Val	Thr	Gly	Ile	Leu	Met	Ile	Ile	Ile	Met	Ser	Phe	Ser	Phe	Thr	Leu	435	440	445	
Ala	Thr	His	Ser	Phe	Arg	Arg	Ser	Val	Val	Lys	Leu	Pro	Ser	Pro	Leu	450	455	460	
His	His	Leu	Ala	Gly	Phe	Asn	Ala	Phe	Trp	Tyr	Ala	His	His	Leu	Leu	465	470	475	480
Val	Leu	Ala	Tyr	Val	Leu	Leu	Val	Val	His	Ser	Tyr	Phe	Ile	Phe	Leu	485	490	495	
Thr	Arg	Glu	Trp	Tyr	Lys	Lys	Thr	Thr	Trp	Met	Tyr	Leu	Ile	Val	Pro	500	505	510	
Val	Leu	Phe	Tyr	Ala	Cys	Glu	Arg	Thr	Ile	Arg	Lys	Val	Arg	Glu	Asn	515	520	525	

52

Asn	Tyr	Arg	Val	Ser	Ile	Val	Lys	Ala	Ala	Ile	Tyr	Pro	Gly	Asn	Val
530						535					540				
Leu	Ser	Leu	His	Met	Lys	Lys	Pro	Pro	Gly	Phe	Lys	Tyr	Lys	Ser	Gly
545					550					555					560
Met	Tyr	Leu	Phe	Val	Lys	Cys	Pro	Asp	Val	Ser	Pro	Phe	Glu	Trp	His
				565					570					575	
Pro	Phe	Ser	Ile	Thr	Ser	Ala	Pro	Gly	Asp	Asp	Tyr	Leu	Ser	Val	His
			580					585					590		
Ile	Arg	Thr	Leu	Gly	Asp	Trp	Thr	Thr	Glu	Leu	Arg	Asn	Leu	Phe	Gly
		595					600					605			
Lys	Ala	Cys	Glu	Ala	Gln	Val	Thr	Ser	Lys	Lys	Ala	Thr	Leu	Ser	Arg
	610					615					620				
Leu	Glu	Thr	Thr	Val	Val	Ala	Asp	Ala	Gln	Thr	Glu	Asp	Thr	Arg	Phe
625					630					635					640
Pro	Lys	Val	Leu	Ile	Asp	Gly	Pro	Tyr	Gly	Ala	Pro	Ala	Gln	Asn	Tyr
				645					650					655	
Lys	Lys	Tyr	Asp	Ile	Leu	Leu	Leu	Ile	Gly	Leu	Gly	Ile	Gly	Ala	Thr
			660					665					670		
Pro	Phe	Ile	Ser	Ile	Leu	Lys	Asp	Leu	Leu	Asn	Asn	Ile	Lys	Ser	Asn
		675					680					685			
Glu	Glu	Val	Glu	Ser	Ile	His	Gly	Ser	Glu	Ile	Gly	Ser	Phe	Lys	Asn
	690					695					700				
Asn	Gly	Pro	Gly	Arg	Ala	Tyr	Phe	Tyr	Trp	Val	Thr	Arg	Glu	Gln	Gly
705					710					715					720
Ser	Phe	Glu	Trp	Phe	Lys	Gly	Val	Met	Asn	Asp	Val	Ala	Glu	Ser	Asp
				725					730					735	
His	Asn	Asn	Ile	Ile	Glu	Met	His	Asn	Tyr	Leu	Thr	Ser	Val	Tyr	Glu
			740					745					750		
Glu	Gly	Asp	Ala	Arg	Ser	Ala	Leu	Ile	Ala	Met	Val	Gln	Ser	Leu	Gln
		755					760					765			
His	Ala	Lys	Asn	Gly	Val	Asp	Ile	Val	Ser	Gly	Ser	Arg	Ile	Arg	Thr
	770					775					780				
His	Phe	Ala	Arg	Pro	Asn	Trp	Arg	Lys	Val	Phe	Ser	Asp	Leu	Ala	Asn
785					790					795					800
Ala	His	Lys	Asn	Ser	Arg	Ile	Gly	Val	Phe	Tyr	Cys	Gly	Ser	Pro	Thr
				805					810					815	
Leu	Thr	Lys	Gln	Leu	Lys	Asp	Leu	Ser	Lys	Glu	Phe	Ser	Gln	Thr	Thr
			820					825					830		
Thr	Thr	Arg	Phe	His	Phe	His	Lys	Glu	Asn	Phe					
		835					840								

<210> 19

<211> 2604

<212> DNA

<213> Arabidopsis thaliana

<220>

<221> CDS

<222> (1)..(2601)

<223> coding" for NADPH oxidase

53

<400> 19

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Met Ser Arg Val Ser Phe Glu Val Ser Gly Gly Tyr His Ser Asp Ala	
1 5 10 15	
gaa gcc gga aac agc gga cca atg agc ggt ggt caa tta cca ccg atc	96
Glu Ala Gly Asn Ser Gly Pro Met Ser Gly Gly Gln Leu Pro Pro Ile	
20 25 30	
tat aaa aaa ccg ggg aac tcc aga ttc act gct gag aac agt cag aga	144
Tyr Lys Lys Pro Gly Asn Ser Arg Phe Thr Ala Glu Asn Ser Gln Arg	
35 40 45	
aca cgt acg gca cca tac gtg gac ctc acg gta gat gta caa gac gat	192
Thr Arg Thr Ala Pro Tyr Val Asp Leu Thr Val Asp Val Gln Asp Asp	
50 55 60	
aca gtc tct gta cat agc ttg aaa atg gaa ggt gga tct agc gtt gaa	240
Thr Val Ser Val His Ser Leu Lys Met Glu Gly Gly Ser Ser Val Glu	
65 70 75 80	
gag agt ccg gag ctt act ttg ctg aaa cga aac cgt ctt gag aag aaa	288
Glu Ser Pro Glu Leu Thr Leu Lys Arg Asn Arg Leu Glu Lys Lys	
85 90 95	
aca acg gtg gtg aaa cgt ttg gcg tct gtt tct cac gag ctt aag cgt	336
Thr Thr Val Val Lys Arg Leu Ala Ser Val Ser His Glu Leu Lys Arg	
100 105 110	
ttg aca tct gtt tct ggt ggt att ggt gga aga aag ccg cct cga ccg	384
Leu Thr Ser Val Ser Gly Gly Ile Gly Gly Arg Lys Pro Pro Arg Pro	
115 120 125	
gct aag tta gac cgg act aaa tcc gcc gcg agt caa gcg ttg aag gga	432
Ala Lys Leu Asp Arg Thr Lys Ser Ala Ala Ser Gln Ala Leu Lys Gly	
130 135 140	
ctt aag ttc att agt aaa acc gac ggt ggc gcc ggt tgg tct gcc gtg	480
Leu Lys Phe Ile Ser Lys Thr Asp Gly Gly Ala Gly Trp Ser Ala Val	
145 150 155 160	
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Glu Lys Arg Phe Asn Gln Ile Thr Ala Thr Thr Gly Gly Leu Leu Leu	
165 170 175	
cgg aca aag ttc ggt gaa tgc ata gga atg act tca aag gat ttt gct	576
Arg Thr Lys Phe Gly Glu Cys Ile Gly Met Thr Ser Lys Asp Phe Ala	
180 185 190	
ttg gaa ctg ttt gat gca ttg gct aga aga agg aat ata aca ggg gaa	624
Leu Glu Leu Phe Asp Ala Leu Ala Arg Arg Arg Asn Ile Thr Gly Glu	
195 200 205	
gtg att gat gga gat caa cta aag gag ttt tgg gaa caa att aat gat	672
Val Ile Asp Gly Asp Gln Leu Lys Glu Phe Trp Glu Gln Ile Asn Asp	
210 215 220	
caa agt ttt gat tct cgg ctt aag aca ttc ttt gac atg gtg gat aaa	720
Gln Ser Phe Asp Ser Arg Leu Lys Thr Phe Phe Asp Met Val Asp Lys	
225 230 235 240	
gat gct gat ggt aga ctt aca gaa gac gaa gtt aga gag ttg gag agt	768
Asp Ala Asp Gly Arg Leu Thr Glu Asp Glu Val Arg Glu Leu Glu Ser	
245 250 255	
ctt gag act ctg ctt ttg caa gcg gca aca cag tct gtg ata aca agt	816
Leu Glu Thr Leu Leu Leu Gln Ala Ala Thr Gln Ser Val Ile Thr Ser	
260 265 270	

54

act ggg gag aga aag aat ctg agt cat atg atg agt cag agg ctt aag	864
Thr Gly Glu Arg Lys Asn Leu Ser His Met Met Ser Gln Arg Leu Lys	
275 280 285	
cct acg ttt aac cgc aac ccg ttg aag cga tgg tac cgt ggt ctt aga	912
Pro Thr Phe Asn Arg Asn Pro Leu Lys Arg Trp Tyr Arg Gly Leu Arg	
290 295 300	
ttc ttc ttg tta gac aac tgg caa aga tgt tgg gtt ata gtg cta tgg	960
Phe Phe Leu Leu Asp Asn Trp Gln Arg Cys Trp Val Ile Val Leu Trp	
305 310 315 320	
ttc ata gtt atg gct ata ctc ttc acc tac aaa tat atc caa tac agg	1008
Phe Ile Val Met Ala Ile Leu Phe Thr Tyr Lys Tyr Ile Gln Tyr Arg	
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Arg Ser Pro Val Tyr Pro Val Met Gly Asp Cys Val Cys Met Ala Lys	
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Gly Ala Ala Glu Thr Val Lys Leu Asn Met Ala Leu Ile Leu Leu Pro	
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Val Cys Arg Asn Thr Ile Thr Trp Leu Arg Asn Lys Thr Arg Leu Gly	
370 375 380	
cgt gtt gtc cca ttt gat gac aat ctc aac ttc cac aag gtt ata gcg	1200
Arg Val Val Pro Phe Asp Asp Asn Leu Asn Phe His Lys Val Ile Ala	
385 390 395 400	
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Val Gly Ile Ile Val Gly Val Thr Met His Ala Gly Ala His Leu Ala	
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tgt gat ttc ccg cgg tta cta cat gca act cca gag gca tat agg cct	1296
Cys Asp Phe Pro Arg Leu Leu His Ala Thr Pro Glu Ala Tyr Arg Pro	
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Leu Arg Gln Phe Phe Gly Asp Glu Gln Pro Lys Ser Tyr Trp His Phe	
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Val Asn Ser Val Glu Gly Ile Thr Gly Leu Val Met Val Leu Leu Met	
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Ala Ile Ala Phe Thr Leu Ala Thr Pro Trp Phe Arg Arg Gly Lys Leu	
465 470 475 480	
aac tat ctt cca gga cca tta aag aaa cta gct agc ttc aat gcc ttc	1488
Asn Tyr Leu Pro Gly Pro Leu Lys Lys Leu Ala Ser Phe Asn Ala Phe	
485 490 495	
tgg tac act cat cat ttg ttt gtc ata gtc tac att ctt ctt gtt gct	1536
Trp Tyr Thr His His Leu Phe Val Ile Val Tyr Ile Leu Leu Val Ala	
500 505 510	
cat gga tac tac ttg tat ctc acc aga gac tgg cac aat aaa acg act	1584
His Gly Tyr Tyr Leu Tyr Leu Thr Arg Asp Trp His Asn Lys Thr Thr	
515 520 525	
tgg atg tat ttg gtg gta cca gtg gtt cta tac gcg tgt gaa agg ttg	1632
Trp Met Tyr Leu Val Val Pro Val Val Leu Tyr Ala Cys Glu Arg Leu	
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ata	aga	gca	ttc	agg	tcg	agc	atc	aag	gcg	gtg	act	att	agg	aaa	gta	1680
Ile	Arg	Ala	Phe	Arg	Ser	Ser	Ile	Lys	Ala	Val	Thr	Ile	Arg	Lys	Val	
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gca	gtt	tat	cca	gga	aac	gtg	ctg	gca	att	cac	ttg	tca	agg	cct	caa	1728
Ala	Val	Tyr	Pro	Gly	Asn	Val	Leu	Ala	Ile	His	Leu	Ser	Arg	Pro	Gln	
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aac	ttc	aaa	tac	aag	agt	ggc	caa	tac	atg	ttt	gtt	aac	tgt	gct	gct	1776
Asn	Phe	Lys	Tyr	Lys	Ser	Gly	Gln	Tyr	Met	Phe	Val	Asn	Cys	Ala	Ala	
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Val	Ser	Pro	Phe	Glu	Trp	His	Pro	Phe	Ser	Ile	Thr	Ser	Ala	Pro	Gln	
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gat	gat	tac	cta	agt	gtt	cac	att	aga	gtt	ctt	ggg	gat	tgg	aca	cga	1872
Asp	Asp	Tyr	Leu	Ser	Val	His	Ile	Arg	Val	Leu	Gly	Asp	Trp	Thr	Arg	
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gct	ctc	aaa	gga	gtc	ttc	tct	gag	gtg	tgt	aag	cca	cca	ccg	gca	gga	1920
Ala	Leu	Lys	Gly	Val	Phe	Ser	Glu	Val	Cys	Lys	Pro	Pro	Pro	Ala	Gly	
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gtt	agt	ggc	ctg	ctt	aga	gcc	gac	atg	ttg	cat	ggc	gca	aac	aac	ccc	1968
Val	Ser	Gly	Leu	Leu	Arg	Ala	Asp	Met	Leu	His	Gly	Ala	Asn	Asn	Pro	
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gac	ttc	ccg	aaa	gtc	ttg	att	gat	ggc	cca	tat	ggc	gca	cca	gca	caa	2016
Asp	Phe	Pro	Lys	Val	Leu	Ile	Asp	Gly	Pro	Tyr	Gly	Ala	Pro	Ala	Gln	
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Asp	Tyr	Lys	Lys	Tyr	Glu	Val	Val	Leu	Leu	Val	Gly	Leu	Gly	Ile	Gly	
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gcc	aca	cca	atg	atc	agt	atc	gtc	aaa	gac	att	gtt	aac	aac	atc	aag	2112
Ala	Thr	Pro	Met	Ile	Ser	Ile	Val	Lys	Asp	Ile	Val	Asn	Asn	Ile	Lys	
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gcc	aag	gaa	caa	gcc	caa	cta	aac	cga	atg	gag	aac	gga	aca	agc	gaa	2160
Ala	Lys	Glu	Gln	Ala	Gln	Leu	Asn	Arg	Met	Glu	Asn	Gly	Thr	Ser	Glu	
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cca	caa	cga	agt	aag	aaa	gag	agt	ttc	agg	acc	cgt	aga	gct	tac	ttc	2208
Pro	Gln	Arg	Ser	Lys	Lys	Glu	Ser	Phe	Arg	Thr	Arg	Arg	Ala	Tyr	Phe	
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tat	tgg	gtt	acg	cgt	gag	caa	ggc	tct	ttc	gat	tgg	ttc	aag	aac	ata	2256
Tyr	Trp	Val	Thr	Arg	Glu	Gln	Gly	Ser	Phe	Asp	Trp	Phe	Lys	Asn	Ile	
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Met	Asn	Glu	Val	Ala	Glu	Arg	Asp	Ala	Asn	Arg	Val	Ile	Glu	Met	His	
		755					760					765				
aac	tat	tgt	aca	agt	gtc	tat	gaa	gaa	ggc	gac	gct	cgt	tcc	gca	ctt	2352
Asn	Tyr	Cys	Thr	Ser	Val	Tyr	Glu	Glu	Gly	Asp	Ala	Arg	Ser	Ala	Leu	
		770				775					780					
ata	cat	atg	ctt	caa	tca	cta	aac	cat	gca	aag	aac	ggc	gtc	gac	att	2400
Ile	His	Met	Leu	Gln	Ser	Leu	Asn	His	Ala	Lys	Asn	Gly	Val	Asp	Ile	
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Val	Ser	Gly	Thr	Arg	Val	Met	Ser	His	Phe	Ala	Lys	Pro	Asn	Trp	Arg	
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56

aat gtt tac aag cgt ata gct atg gat cat cct aac acc aaa gtt gga 2496
 Asn Val Tyr Lys Arg Ile Ala Met Asp His Pro Asn Thr Lys Val Gly
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gtg ttt tac tgt gga gca cca gca ttg aca aag gag cta agg cat cta 2544
 Val Phe Tyr Cys Gly Ala Pro Ala Leu Thr Lys Glu Leu Arg His Leu
 835 840 845

gct tta gat ttc acc cac aaa aca agc acc aga ttc tcc ttc cac aaa 2592
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<213> Arabidopsis thaliana

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 35 40 45

Thr Arg Thr Ala Pro Tyr Val Asp Leu Thr Val Asp Val Gln Asp Asp
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Thr Val Ser Val His Ser Leu Lys Met Glu Gly Gly Ser Ser Val Glu
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Glu Ser Pro Glu Leu Thr Leu Leu Lys Arg Asn Arg Leu Glu Lys Lys
 85 90 95

Thr Thr Val Val Lys Arg Leu Ala Ser Val Ser His Glu Leu Lys Arg
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Leu Thr Ser Val Ser Gly Gly Ile Gly Gly Arg Lys Pro Pro Arg Pro
 115 120 125

Ala Lys Leu Asp Arg Thr Lys Ser Ala Ala Ser Gln Ala Leu Lys Gly
 130 135 140

Leu Lys Phe Ile Ser Lys Thr Asp Gly Gly Ala Gly Trp Ser Ala Val
 145 150 155 160

Glu Lys Arg Phe Asn Gln Ile Thr Ala Thr Thr Gly Gly Leu Leu Leu
 165 170 175

Arg Thr Lys Phe Gly Glu Cys Ile Gly Met Thr Ser Lys Asp Phe Ala
 180 185 190

Leu Glu Leu Phe Asp Ala Leu Ala Arg Arg Arg Asn Ile Thr Gly Glu
 195 200 205

Val Ile Asp Gly Asp Gln Leu Lys Glu Phe Trp Glu Gln Ile Asn Asp
 210 215 220

Gln Ser Phe Asp Ser Arg Leu Lys Thr Phe Phe Asp Met Val Asp Lys
 225 230 235 240

Asp Ala Asp Gly Arg Leu Thr Glu Asp Glu Val Arg Glu Leu Glu Ser
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Thr	Gly	Glu	Arg	Lys	Asn	Leu	Ser	His	Met	Met	Ser	Gln	Arg	Leu	Lys
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Pro	Thr	Phe	Asn	Arg	Asn	Pro	Leu	Lys	Arg	Trp	Tyr	Arg	Gly	Leu	Arg
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Phe	Phe	Leu	Leu	Asp	Asn	Trp	Gln	Arg	Cys	Trp	Val	Ile	Val	Leu	Trp
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Phe	Ile	Val	Met	Ala	Ile	Leu	Phe	Thr	Tyr	Lys	Tyr	Ile	Gln	Tyr	Arg
				325					330					335	
Arg	Ser	Pro	Val	Tyr	Pro	Val	Met	Gly	Asp	Cys	Val	Cys	Met	Ala	Lys
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Gly	Ala	Ala	Glu	Thr	Val	Lys	Leu	Asn	Met	Ala	Leu	Ile	Leu	Leu	Pro
		355					360					365			
Val	Cys	Arg	Asn	Thr	Ile	Thr	Trp	Leu	Arg	Asn	Lys	Thr	Arg	Leu	Gly
	370					375					380				
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Val	Gly	Ile	Ile	Val	Gly	Val	Thr	Met	His	Ala	Gly	Ala	His	Leu	Ala
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Cys	Asp	Phe	Pro	Arg	Leu	Leu	His	Ala	Thr	Pro	Glu	Ala	Tyr	Arg	Pro
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Leu	Arg	Gln	Phe	Phe	Gly	Asp	Glu	Gln	Pro	Lys	Ser	Tyr	Trp	His	Phe
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Trp	Tyr	Thr	His	His	Leu	Phe	Val	Ile	Val	Tyr	Ile	Leu	Leu	Val	Ala
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His	Gly	Tyr	Tyr	Leu	Tyr	Leu	Thr	Arg	Asp	Trp	His	Asn	Lys	Thr	Thr
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Trp	Met	Tyr	Leu	Val	Val	Pro	Val	Val	Leu	Tyr	Ala	Cys	Glu	Arg	Leu
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Asp	Asp														

58

Val Ser Gly Leu Leu Arg Ala Asp Met Leu His Gly Ala Asn Asn Pro
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 Asp Phe Pro Lys Val Leu Ile Asp Gly Pro Tyr Gly Ala Pro Ala Gln
 660 665 670
 Asp Tyr Lys Lys Tyr Glu Val Val Leu Leu Val Gly Leu Gly Ile Gly
 675 680 685
 Ala Thr Pro Met Ile Ser Ile Val Lys Asp Ile Val Asn Asn Ile Lys
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 Ala Lys Glu Gln Ala Gln Leu Asn Arg Met Glu Asn Gly Thr Ser Glu
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 Pro Gln Arg Ser Lys Lys Glu Ser Phe Arg Thr Arg Arg Ala Tyr Phe
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 Tyr Trp Val Thr Arg Glu Gln Gly Ser Phe Asp Trp Phe Lys Asn Ile
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 Met Asn Glu Val Ala Glu Arg Asp Ala Asn Arg Val Ile Glu Met His
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 Asn Tyr Cys Thr Ser Val Tyr Glu Glu Gly Asp Ala Arg Ser Ala Leu
 770 775 780
 Ile His Met Leu Gln Ser Leu Asn His Ala Lys Asn Gly Val Asp Ile
 785 790 795 800
 Val Ser Gly Thr Arg Val Met Ser His Phe Ala Lys Pro Asn Trp Arg
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<223> coding for NADPH oxidase

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 Ala Thr Thr Ser Asn Tyr Tyr Gly Glu Asp Glu Pro Tyr Val Glu Ile
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59

acg cta gat atc cac gac gat tcc gtc tcc gtg tac ggc ttg aag tca	192
Thr Leu Asp Ile His Asp Asp Ser Val Ser Val Tyr Gly Leu Lys Ser	
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Pro Asn His Arg Gly Ala Gly Ser Asn Tyr Glu Asp Gln Ser Leu Leu	
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Arg Gln Gly Arg Ser Gly Arg Ser Asn Ser Val Leu Lys Arg Leu Ala	
85 90 95	
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Ser Ser Val Ser Thr Gly Ile Thr Arg Val Ala Ser Ser Val Ser Ser	
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Ser Ser Ala Arg Lys Pro Pro Arg Pro Gln Leu Ala Lys Leu Arg Arg	
115 120 125	
tcg aaa tct aga gca gag cta gct ctc aaa ggt ctt aaa ttc atc acc	432
Ser Lys Ser Arg Ala Glu Leu Ala Leu Lys Gly Leu Lys Phe Ile Thr	
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aag act gat ggt gtc act ggt tgg cct gaa gtt gag aaa cgg ttt tat	480
Lys Thr Asp Gly Val Thr Gly Trp Pro Glu Val Glu Lys Arg Phe Tyr	
145 150 155 160	
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Val Met Thr Met Thr Asn Asn Gly Leu Leu His Arg Ser Arg Phe Gly	
165 170 175	
gaa tgt ata ggg atg aaa tcg acg gag ttt gcg ttg gca ttg ttc gat	576
Glu Cys Ile Gly Met Lys Ser Thr Glu Phe Ala Leu Ala Leu Phe Asp	
180 185 190	
gct tta gcg agg agg gaa aac gta agc gga gat tca ata aac atg aat	624
Ala Leu Ala Arg Arg Glu Asn Val Ser Gly Asp Ser Ile Asn Met Asn	
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Glu Leu Lys Glu Phe Trp Lys Gln Ile Thr Asp Gln Asp Phe Asp Ser	
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Arg Leu Arg Thr Phe Phe Ala Met Val Asp Lys Asp Ser Asp Gly Arg	
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Leu Asn Glu Ala Glu Val Arg Glu Ile Ile Thr Leu Ser Ala Ser Ala	
245 250 255	
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Asn Glu Leu Asp Asn Ile Arg Arg Gln Ala Asp Glu Tyr Ala Ala Leu	
260 265 270	
att atg gaa gaa ctc gat cct tat cat tat gga tac atc atg ata gag	864
Ile Met Glu Glu Leu Asp Pro Tyr His Tyr Gly Tyr Ile Met Ile Glu	
275 280 285	
aat ctc gag ata ctt cta ttg caa gcg ccg atg cag gat gtg aga gat	912
Asn Leu Glu Ile Leu Leu Leu Gln Ala Pro Met Gln Asp Val Arg Asp	
290 295 300	
gga gag agt aag aag cta agc aag atg cta agt cag aat ctc atg gtt	960
Gly Glu Ser Lys Lys Leu Ser Lys Met Leu Ser Gln Asn Leu Met Val	
305 310 315 320	

60

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Phe	Leu	Phe	Asp	Asn	Trp	Lys	Arg	Val	Trp	Val	Met	Ala	Leu	Trp	Ile	
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Gly	Ala	Met	Ala	Gly	Leu	Phe	Thr	Trp	Lys	Phe	Met	Glu	Tyr	Arg	Lys	
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Arg	Ser	Ala	Tyr	Glu	Val	Met	Gly	Val	Cys	Val	Cys	Ile	Ala	Lys	Gly	
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Ala	Ala	Glu	Thr	Leu	Lys	Leu	Asn	Met	Ala	Met	Ile	Leu	Leu	Pro	Val	
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Cys	Arg	Asn	Thr	Ile	Thr	Trp	Leu	Arg	Thr	Lys	Thr	Lys	Leu	Ser	Ala	
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Ile	Val	Pro	Phe	Asp	Asp	Ser	Leu	Asn	Phe	His	Lys	Val	Ile	Ala	Ile	
			420					425					430			
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Gly	Ile	Ser	Val	Gly	Val	Gly	Ile	His	Ala	Thr	Ser	His	Leu	Ala	Cys	
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gat	ttc	ccc	cga	ctg	ata	gct	gca	gac	gaa	gat	cag	tat	gag	cca	atg	1392
Asp	Phe	Pro	Arg	Leu	Ile	Ala	Ala	Asp	Glu	Asp	Gln	Tyr	Glu	Pro	Met	
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Ser	Val	Glu	Gly	Val	Thr	Gly	Ile	Gly	Met	Val	Val	Leu	Met	Thr	Ile	
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cct	gga	cca	ctg	aag	aaa	ata	aca	ggc	ttc	aat	gcc	ttc	tgg	tac	tct	1584
Pro	Gly	Pro	Leu	Lys	Lys	Ile	Thr	Gly	Phe	Asn	Ala	Phe	Trp	Tyr	Ser	
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His	His	Leu	Phe	Val	Ile	Val	Tyr	Ser	Leu	Leu	Val	Val	His	Gly	Phe	
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Tyr	Leu	Met	Val	Pro	Val	Val	Leu	Tyr	Leu	Cys	Glu	Arg	Leu	Ile	Arg	
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Ala	Phe	Arg	Ser	Ser	Val	Glu	Ala	Val	Ser	Val	Leu	Lys	Val	Ala	Val	
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62

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64

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<212> DNA

<213> Artificial sequence

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<210> 24

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<212> DNA

<213> Artificial sequence

<220>

<223> Description of the artificial sequence:
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<400> 24

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We claim

1. A method for generating or increasing the resistance to at least one pathogen in plants, which comprises the following operating steps
 - a) reduction of the protein quantity, activity or function of an NADPH oxidase in a plant or a tissue, organ, part or cell thereof, and
 - b) selection of the plants in which - in contrast or in comparison with the starting plant - the resistance to at least one pathogen exists or is increased.
2. The method according to claim 1, wherein the NADPH oxidase is encoded by
 - a) polypeptide sequences comprising a sequence as shown in SEQ ID NO: 2, 4, 6, 8, 10, 12, 14, 16, 18, 20 or 22, or
 - b) polypeptide sequences of a functional equivalent of a polypeptide comprising a sequence as shown in SEQ ID NO: 2, 4, 6, 8, 10, 12, 14, 16, 18, 20 or 22.
3. The method according to claim 2, wherein the functional equivalent has at least 50% homology with one of the polypeptides as shown in SEQ ID NO: 2, 4, 6, 8, 10, 12, 14, 16, 18, 20 or 22.
4. The method according to any of claims 1 to 3, wherein the reduction of the protein quantity, activity or function of an NADPH oxidase is ensured by applying a method selected from the group consisting of
 - a) introducing a double-stranded NADPH oxidase RNA nucleic acid sequence or (an) expression cassette(s) ensuring its expression,
 - b) introducing an NADPH oxidase antisense nucleic acid sequence or an expression cassette ensuring its expression,

Seq. + drawings

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- c) introducing an NADPH oxidase antisense nucleic acid sequence in combination with a ribozyme or an expression cassette ensuring its expression,
 - 5 d) introducing NADPH oxidase sense nucleic acid sequences for inducing a cosuppression or an expression cassette ensuring their expression,
 - 10 e) introducing DNA- or protein-binding factors against NADPH oxidase genes, RNAs or proteins or an expression cassette ensuring their expression,
 - 15 f) introducing viral nucleic acid sequences and expression constructs bringing about the degradation of NADPH oxidase RNA, or an expression cassette ensuring their expression,
 - 20 g) introducing constructs for inducing a homologous recombination at endogenous NADPH oxidase genes, and
 - h) introducing mutations into an endogenous NADPH oxidase gene.
5. The method according to any of claims 1 to 4, comprising
- 25 (i) the stable transformation of a plant cell with a recombinant expression cassette comprising, in functional linkage with a promoter which is active in plants, a nucleic acid sequence encoding
- 30 a) a double-stranded NADPH oxidase RNA ribonucleic acid sequence or
- b) an NADPH oxidase antisense nucleic acid sequence or
- 35 c) an NADPH oxidase antisense nucleic acid sequence in combination with a ribozyme or
- d) an NADPH oxidase sense nucleic acid sequence for inducing a cosuppression or
- 40 e) DNA- or protein-binding factors against NADPH oxidase genes, RNAs or proteins
- 45 f) viral nucleic acid sequences which bring about the degradation of NADPH oxidase RNA,

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- (ii) regeneration of the plant from the plant cell, and
- (iii) expression of said nucleic acid sequence in such a quantity and for such a time as suffices for generating or increasing a pathogen resistance in said plant.
- 5
6. The method according to any of claims 1 to 5, wherein the pathogen is selected from the group consisting of bacteria, fungi, insects, viruses and nematodes.
- 10
7. The method according to any of claims 1 to 6, wherein the pathogen is selected from the group of the fungi consisting of Plasmodiophoromycota, Oomycota, Ascomycota, Chytridiomycetes, Zygomycetes, Basidiomycota and Deuteromyceten.
- 15
8. The method according to any of claims 1 to 7, wherein the plant is selected from among the monocotyledonous and dicotyledonous plants.
- 20
9. The method according to any of claims 1 to 8, wherein the plant is selected from the group of the monocotyledonous plants consisting of wheat, oats, millet, barley, rye, maize, rice, buckwheat, sorghum, triticale, spelt, linseed or sugar cane.
- 25
10. A double-stranded RNA molecule for reducing the expression of an NADPH oxidase protein comprising
- 30
- a) a sense RNA strand comprising at least one ribonucleotide sequence which is essentially identical to at least part of the sense RNA transcript of a nucleic acid sequence encoding an NADPH oxidase, and
- 35
- b) an antisense RNA strand which is essentially complementary to the RNA sense strand of a).
11. The double-stranded RNA molecule according to claim 10, wherein the two RNA strands of the double-stranded RNA are bonded covalently with one another.
- 40
12. The double-stranded RNA molecule according to either of claims 10 or 11, wherein one of the two RNA strands is encoded by at least a part of the nucleic acid sequence encoding an NADPH oxidase sequence as shown in SEQ ID NO: 1, 3, 5, 7, 9, 11, 13, 15, 17, 19 or 21 or a functional equivalent thereof.
- 45

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13. A transgenic expression cassette comprising, in functional linkage with a promoter which is functional in plant organisms, a nucleic acid sequence encoding a double-stranded RNA molecule according to one of Claims 10 to 12.
- 5
14. A transgenic expression cassette comprising at least a part of a nucleic acid sequence encoding an NADPH oxidase as shown in SEQ ID NO: 1, 3, 5, 7, 9, 11, 13, 15, 17, 19 or 21 or a functional equivalent thereof, where said nucleic acid sequence is linked functionally in antisense orientation with a promoter which is functional in plant organisms.
- 10
15. The transgenic expression cassette according to claim 13 or 14, wherein the promoter which is functional in plants is a pathogen-inducible promoter.
- 15
16. A transgenic vector comprising an expression cassette according to any of claims 13 to 15.
- 20
17. A transgenic organism comprising a double-stranded RNA molecule according to any of claims 10 to 12, an expression cassette according to any of claims 13 to 15 or a vector according to claim 16.
- 25
18. The transgenic organism according to claim 17, selected from the group consisting of bacteria, yeasts, animals and plants.
19. The transgenic organism according to claim 17 or 18, selected from the group of the plants consisting of wheat, oats, millet, barley, rye, maize, rice, buckwheat, sorghum, triticale, spelt, linseed, sugar cane, oilseed rape, canola, cress, Arabidopsis, cabbages, soybeans, alfalfa, pea, beans, peanut, potato, tobacco, tomato, egg plant, capsicum, sunflower, Tagetes, lettuce, Calendula, melon, pumpkin/squash and zucchini.
- 30
- 35
20. A tissue, organ, part, cell, cell culture or propagation material derived from a transgenic organism according to either of claims 18 or 19.
- 40

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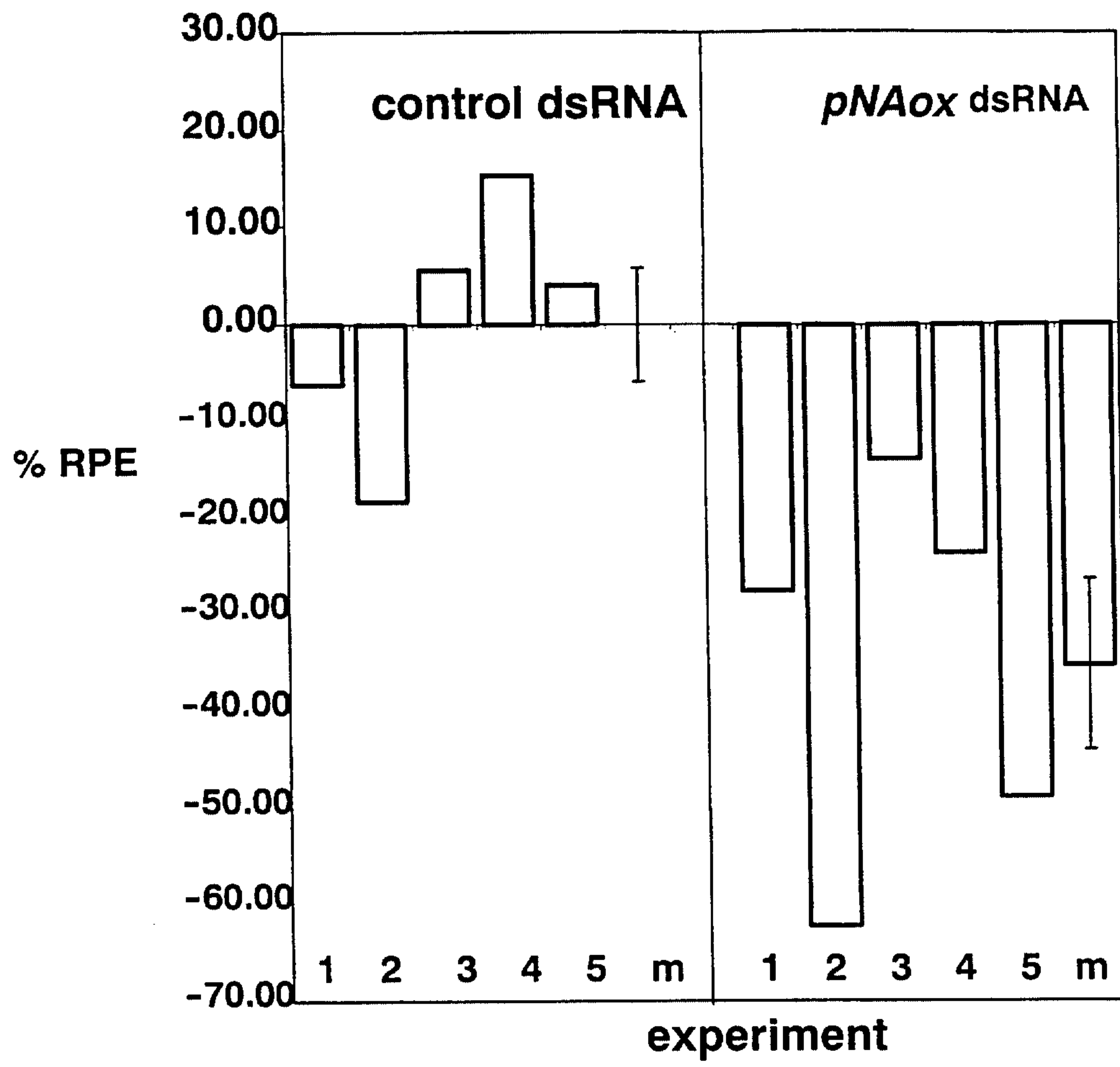


Fig. 1