The International Treaty
ON PLANT GENETIC RESOURCES FOR FOOD AND AGRICULTURE

Full Project Proposal

Third Call for Proposals under the Benefit-sharing Fund

Deadline for submitting full project proposal: 5th of December 2014
at Treaty-Fund@fao.org and PGRFA-Treaty@fao.org

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PROJECT PROPOSAL COVER SHEET

Project No. _____ (*For Treaty use. Do not write anything here)*

Project Title: **MARKER ASSISTED SELECTION FOR POTATO GERMPLASM ADAPTED TO BIOTIC AND ABIOTIC STRESSES CAUSED BY GLOBAL CLIMATE CHANGE**

Project duration: **36 months**

Target crops: **Potato, including Native Potato species**

Targeted developing country/ies PERU, ECUADOR, VENEZUELA

Other Contracting Party/ies involved SPAIN (SUBCONTRACTOR)

Project geographic extension (km²) 2.945.000

Total requested funding: **497.585 US$**

Total co-funding: **522.000 US$**

Please select the type of project you are applying for:

☐ Single-country Immediate Action Project (Window 2)

☐ Multi-country Immediate Action Programme (Window 2)

☐ Single-country Co-development and Transfer of Technology project (Window 3)

☒ Multi-country Co-development and Transfer of Technology project (Window 3)

Applicant

Name of Organization: **UNIVERSIDAD NACIONAL AGRARIA LA MOLINA (UNALM) – Instituto de Biotecnología (IBT)**

Type of organization **UNIVERSITY**

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GENERAL REQUIREMENTS

These guidelines have been prepared to support applicants in the development of full project proposals. They describe the requirements that all applicants should adhere to when developing their full project proposal.

Please make sure you read these guidelines carefully before proceeding to fill in the Project Proposal Form. The full proposal should be prepared taking into account the thematic focus of the Third Call for Proposals, including in particular, the rationale, scope and expected outputs for each Window and sub-Window.

Project proposals must be clear and realistic on the problem to be addressed and objectives to be achieved. Project objectives have to fit in the thematic focus of the call and ultimately contribute to food security and poverty alleviation. Project objectives have to be logically interlinked with the planned activities, outputs and expected outcomes. The objectives and outputs have to be feasible in terms of duration and resources requested. The information to be provided in each section has to be focused and straightforward, qualitatively and quantitatively measurable in terms of what will be done, with what purpose, who will be involved in the activities to be implemented, who and how many will directly and indirectly benefit from the implementation of the project. A good full proposal will have a sound, clear and logically linked methodology of implementation and management.

The full project proposal should contain **no more than fifteen (15) pages** of text (Appendixes, table of contents and cover sheets excluded). The number of pages allocated to each section is a guide. The information required can be less but not more than the number of pages stipulated. All Appendixes should be duly filled in according to the provided guidelines as they form an integral part of the full project proposal. Project proposals lacking even one Appendix, will be excluded from the selection process. The Appendixes will be provided to you in separate files together with the present document.

When submitting the full project proposal, additional attachments (endorsement letters, funding commitments, certification of the status of the organization) can also be submitted with the main proposal.

Please ensure that the project proposal and all attachments are legible in **Times New Roman 12** and provided in two formats (**pdf and word**). Make sure the signature of the project coordinator is put on the signature page.

The project proposal, if approved for funding by the Bureau of the Sixth Session of the Governing Body, will form an integral part of the contractual agreement (Letter of Agreement) that will be signed with each applicant organization of the approved projects.

SECTION A: EXECUTIVE SUMMARY

1. Executive summary

The project belongs to Window 3: Support to the co-development and Technology Transfer involving a Consortium of 4 partners from 3 Andean countries and Spain. It addresses Potato including Native Potato species which play a key role for food security and subsistence of Andean farmers. Abiotic stresses and related biotic stresses caused by climate change represent a critical limitation and a mayor threat for sustainable agriculture and food security. It is necessary to develop new cultivars with tolerance to these stresses by exploiting the existing biodiversity of species. In this project we will characterize in part novel, yet unexploited potato germplasm from the Andes and identify accessions which are adapted to these threats of climate change. Based on this information we will develop molecular markers which can be used for potato breeding of new improved potato cultivars with elevated stress tolerance levels for sustainable agriculture.

Phenotypic evaluations of these accessions for resistance or tolerance to abiotic and associated biotic stresses will be performed in field trials and bio-assays. The traits for evaluation include abiotic stresses *cold, drought and heat as well as* the major fungal pathogen *Phytopthora infestans*. The identification of tolerant genotypes will provide directly recommendations to farmers for cultivation of these varieties in environments with adverse agro-climatical conditions, and represent at the same time valuable material for the breeding of improved potato varieties.

On the other hand we will detect candidate genes (CG) for resistance or tolerance to these stresses using different up to date molecular tools. These include RNAseq, an in silico mining approach of known genes and RAD sequencing. We will analyse the allelic variation of these CG and determine the effect of specific alleles or allele combinations in the materials through amplicon sequencing and association mapping by linking the phenotypic data of the previous evaluations with the obtained molecular data. CG detection and analyses of alleles will be also performed using a random approach, known as RAD sequencing. The results will allow to develop markers for marker assisted selection, which can be applied to speed up conventional potato breeding programs. Results of individual CG will be extended to multiple CG and combined for multiple traits through Model building with the practical aim to assign parental breeding values and predict progeny performances in order to realize optimized crosses.

Pre-breeding activities by means of crossings and evaluations of resulting progenies will be performed to combine favourable characteristics and to improve adaptation to climate change, supported by the developed markers and models.

All Project results and Products (breeding clones) will be disseminated and transferred between partners, but also to farmer associations, to the scientific community, to breeders and to gene bank curators through numerous dissemination and transfer actions. A Project WEB page with an integrated Knowledge base will be established containing all project results and external links. The molecular markers and Models for analysing stress adaptation in potatoes can be used for efficient marker assisted breeding in potato and related species.

SECTION B: PROJECT DESCRIPTION AND CONTENTS

2.1. Problem definition

The effects of global climate change such as heat, coldness, drought or flooding are likely to threaten most crop species. Moreover, changes in the pathogen spectrums affecting a crop can be expected and has been already observed for *Phytopthora infestans*, the most important potato pathogen. In Peru this pathogen is now affecting potato at altitudes where never occurred (3900-4300 masl) posing a risk to loss native potatoes and its wild relatives (Fernández-Northcote, E.N. Comunicación Personal). In Ecuador significative changes in amount and distribution of rain has also provoked increased incidence of *P. infestans* and other pests like nematodes (Perez et al., 2010; Seo and Mendelsohn, 2008; Seo, 2011). In Venezuela the most probable scenarios in agriculture production points to reduction due to either flooding or prolonged dry seasons that the country has experienced in recent years, increased temperatures that affects crop yield as well as pathogen and pests dynamics, like *P. infestans*, among others.

Alleviating measures include development and adoption of new varieties adapted to biotic and biotic stresses (Martelo, 2009). Both sources of stress, biotic and abiotic, are included as research targets under National Strategies in Peru, Ecuador and Venezuela (Estrategia Nacional de Cambio Climático del Peru, ENCC, Decreto Supremo N° 031 – 2008 – AG published 28/06/2008); (http://www.pacc-ecuador.org/cambio-climatico); (Ministerio de Ciencia y Tecnología, 2005); (Levis, 2009; Vargas, 2009). It is necessary to rapidly develop these new cultivars which are adapted and resilient to these locally variable threats by applying marker assisted selection (MAS) or genetic transformations based on useful candidate genes.

Potato (*Solanum tuberosum*) ranks as the world's third most important food crop after wheat and rice (maize is used predominantly as fodder) outstripping all other food crops in developing countries in terms of growth in production area (CIP, 2013). Potato provides a significant contribution to the global food supply and is one of the principal sources of food, productive employment above 3000 m above sea level (MINAGRI, 2014), and income for marginalized citizens and vulnerable small-scale farmers of developing countries in the Andean region and around the world (CIP, 2013) (Devaux et al., 2010; Mancero, 2007; Monteros, 2011).

Particularly, potato plays a key role for food security and subsistence of Andean farmers, as recognized by the initiatives and plans in food security and nutrition of the governments of the Andean Community.

Most of the actually cultivated potato species are not adapted to the threats of climate change, but large germplasm resources in form of native and wild *Solanum* species exist which carry important genes for resistance or tolerance to different stresses.

Large part of the genetic variation is located in the Andean regions of Peru and Bolivia. The aim of this project is to characterize cultivated and wild germplasm with respect to resistance and tolerance to different biotic and abiotic stresses and exploit it as fast as possible through modern breeding to obtain new potato varieties adapted to climate change for sustainable agriculture.

Genomic studies offer the possibility to characterize germplasm efficiently at the molecular level and to accelerate considerably breeding programmes. The detection of candidate genes for useful traits offers the possibility to apply them – after developing the corresponding markers – in marker assisted selection (MAS) within breeding programmes. The survey of allelic diversity of such genes within cultivated and wild species and analyses of their particular effects permits to select the most efficient allele combinations for these purposes. Within this project we want to identify useful candidate genes for different biotic and abiotic stresses using molecular tools, characterize the allelic variation of this germplasm and use the markers in marker assisted breeding, which has not been approached before for the Andean region.

2.2. Project objectives: Overall and specific objectives

The **General Objective** consists of identifying potato accessions adapted to biotic and abiotic threats of climate change, to identify the underlying candidate genes for developing molecular markers and models, which will speed up the breeding of improved and adapted potato cultivars for sustainable agriculture.

In order to meet this general objective the following **Specific Objectives** are envisaged:

1. Evaluation of potato accessions (cultivars, breeding clones, native and wild *Solanum* species) for resistance or tolerance to abiotic and biotic stresses related to global climate change.
2. Detection of useful candidate genes (CG) for abiotic and associated biotic stresses applying different molecular Tools.
3. Molecular characterization of the allelic variation in these CG and determination of allelic composition in the evaluated accessions.
4. Association mapping to detect the effects of specific CG alleles or CG allele combinations on the tolerance levels of the analysed stresses, development of molecular markers for Marker-Assisted Selection and Model building to assign parental breeding values and predict progeny performances.
5. Pre-breeding activities to combine favourable characteristics and to improve adaptation to climate change applying the developed markers and models.
6. Dissemination and Transfer of Project results and Products (accessions and breeding clones).

2.3 Targeted outputs, activities and related methodology of implementation

Participants:

The R&D activities will be carried out jointly by four public institutions:

P1. IBT - Instituto de Biotecnología de la Universidad Nacional Agraria La Molina (Lead Institution, Peru)

P2. INIAP - National Agriculture Research Institute (Ecuador)

P3. ULA: Universidad de Los Andes (Mérida, Venezuela) - Laboratorio de Biodiversidad y Variabilidad Molecular, Instituto Jardín Botánico de Mérida

P4: NEIKER - Basque Institute for Research and Development in Agriculture, Spain (Subcontracted)

Specific Objective 1: Phenotypic evaluation of the germplasm working collection

Target Output 1: Andean potato varieties and accessions including native potato species with resistance or tolerance to abiotic and associated biotic stresses related to global climate change identified, recommended for cultivation under adverse conditions and used for cultivation and breeding.

Activity	Project outputs	Targeted outputs (Deliverable)	Due date
A1.1	Results on evaluations of drought, cold and heat tolerance of the accessions through field trials and bio-assays.	**D1.1a,b:** Evaluation Data & Recommended LIST of accessions with tolerance to different abiotic stresses for cultivation & breeding	Months* 12 and 24
A1.2	Results of evaluation assays for resistances to *P. infestans* in the	**D1.2a,b:** Evaluation Data & Recommended LIST of accessions	Months* 12 and

	working collection.	with resistance to *P. infestans* for cultivation & breeding	24

* ***First results and final results, respectively.***

Activities

A1.1: To carry out field trials to evaluate agronomic performance, resistance or tolerance to abiotic stress factors: drought, cold, heat, and to identify promising, adapted accessions.

A1.2 To carry out field trials and bioassays to evaluate resistance to *P. infestans* and to identify resistant accessions

Methodology

Activity 1.1: Evaluation of resistance or tolerance to abiotic stress factors: drought, cold, heat.

Partners **P1, P2, P3** will perform field trials as specified in **Table 1** at locations with different stress conditions and at locations without stress (control), as well as in bio-assays under controlled conditions, using standard methodology. A block design of single plant plots will be implemented, with four repetitions. The partners will record general agronomic performance, yield, tuber number, tuber weight and starch content (or specific gravity) under normal and stressed conditions. In order to calculate stress tolerance levels, absolute and relative stress-induced yield losses will be computed. For combining the data from different trials, all values will be expressed as relative values with respect to the trial mean (100%).

Activity 1.2: Evaluation of resistance or tolerance to late blight (*Phytophthora infestans*)

Partners **P1, P2, P3** will evaluate also the incidence of *P. infestans* in the materials of the field trials at locations with high infection pressure. AUDPC values (Andrivon et al. 2006) to measure the disease progress after infection with *P. infestans* will be measured. In addition, Phytophthora resistance will be determined according to Michalska et al. (2011) in bio-assays using detached leaflets to complete the evaluations.

Specific Objective 2: Detection of useful candidate genes (CG) for abiotic and biotic stresses and Analysis of the allelic variation in these CG

Target Output 2: Useful candidate genes for abiotic stress and associated biotic stress tolerance identified applying RNAseq, in silico Mining and RAD sequencing & existing allelic variation for these CG in the evaluated accessions determined.

Activity	Project outputs	Targeted outputs (Deliverable)	Due date
A2.1	Results of CG analyses derived from RNAseq sequences for drought, cold and heat tolerance.	**D2.1a,b:**List of new candidate genes for abiotic and related biotic stress resistance derived from RNAseq.	Months* 12 and 24
A2.2	Results of *in silico* mining to detect published candidate genes for tolerances to abiotic stresses drought, cold, heat and resistance to biotic stresses. Identification of homologues in potato.	**D2.2a,b:** List of potato CG derived from in silico mining of published candidate genes for abiotic and related biotic stress resistance.	Months* 12 and 24
A2.3	CG sequences and Amplicon primers. Results of allelic variation of CG and allele composition of the accessions derived from Amplicon sequencing.	**D2.3a,b:** List of CG Sequences and functional primers for obtaining CG amplicons. For each CG LIST of SNP/alleles in the collection and CG allele composition of each accession.	Months* 12 and 18

A2.4	Results of CG extractions derived from RAD sequences, allelic variation of CG and allele composition of the accessions	**D2.4a,b:** LIST of CG extracted from RAD tags and their biological meaning. LIST of SNP/alleles in the collection and CG allele composition of each accession.	Months* 18 and 24

Activities
A2.1: To detect useful CG applying RNA-Seq in stressed and unstressed, susceptible and tolerant genotypes
A2.2: To detect useful CG by analyzing published known genes from potato and other species.
A2.3: To perform successfully Amplicon sequencing of CG in a set of 210 accessions from the field trials (WP1) and to analyze the allelic variation in these Candidate genes.
A2.4: To perform successfully RAD sequencing in this set of 210 accessions, to extract additional CG with a relevant biological meaning and to analyze the allelic variation in the extracted CG.

Methodology

Candidate genes will be detected initially using different molecular approaches and analysed for their allelic varibility:

Activity 2.1 Library construction and RNA-Seq for CG detection
Partner **P4** will perform this task, applying the following workflow: RNA will be extracted from selected susceptible and resistant genotypes cultivated under stressed (cold, drought, heat) and unstressed conditions. Barcoded strand-specific RNA-seq libraries will be constructed by partner **P4** according to Merrick et al (2013) for each sample and multiplexed for sequencing using the Ion Torrent PGM platform. Since a reference genome is available for potato, we will align RNA-seq reads using standard protocols to identify differences between treatments in sense and antisense transcript expression, splicing and allele-specific expression. Homology searches (via NCBI) will detect potential candidate genes with a relevant biological meaning.
Activity 2.2 Analysis of known candidate genes for biotic and abiotic stresses.
Partner 4 will perform in silico mining of sequence databases and publications in order to detect published candidate genes in potato but also in other species. In this latter case the potato homologs will be identified through BLAST searches against the Potato whole genome sequence.
Activity 2.3: Analyses of CG by Amplicon Sequencing (CG driven approach)
Partners **P1**, **P2** and **P3** will extract DNA from 70 accessions each, which are used for phenotyping in the field trials of A1 and send them to Partner P4 (210 accessions in total).

Partner **P4** will design for each identified candidate gene from Activities 2.1 and 2.2 appropriate primers in conserved exon regions based on the sequence information and homology searches and validate them initially in a small subset of genotypes by producing distinct and clear amplification products. Validated primers with a common extension will be used to produce amplicons in the set of 210 genotypes. The bands will be re-amplified via PCR in each genotype using specific barcode primers, which will allow to distinguish the origin (i.e: genotype) of each sample.
After verifying the quality of these final amplification products in gels, aliquots of each sample will be mixed in equal concentrations, and this mix of sample DNAs will be sent for sequencing using the "ION TORRENT Amplicon Sequencing" technique (Life Sciences).
After receiving millions of sequence reads from the Sequencing Platform (A2.3), Partner P4 will order and separate them by candidate gene and genotype. The number of different SNPs and patterns (alleles) which exist in the collection and their frequencies will be determined, as well as their frequencies in the population and the allele composition in each genotype of the collection.
Activity 2.4: RAD sequencing for CG detection and analyses (random approach).

RAD (Restriction site associated DNA marker) sequencing and similar techniques (GBS, GWAS, Genomic Selection) allow potentially to identify many hundreds of CG at a time by screening the whole genome. We will apply a novel, modified RAD sequencing approach based on cDNA templates in order to capture the coding regions of the genome.
For this purpose Partners **P1**, **P2** and **P3** will extract each also total RNA from the same 70 accessions of the field trials and send them to Partner P4.
P4 will extract from each sample mRNA, perform reverse transcription and produce ds-cDNA samples. Restriction fragments will be produced by digesting with Ase + Taq. Following size selection and purification, appropriate adapters will be ligated. After re-amplification with barcoded fusion primers to identify later on the original genotype, the samples will be mixed in equal amounts and sent for Amplicon Sequencing using this time the novel ION PROTON technology (10 Gb chip).

The millions of obtained RAD sequences will be analyzed in a similar way as in A2.3. Restriction fragments will be extracted by homology searches and the allelic variability in terms of SNPs and Patterns (alleles) which exist in the collection will be determined for each extracted CG, as well as the allele composition in each genotype of the collection. Homology searches of RAD markers will be performed in order to identify potential CG with a relevant biological function for explaining stress tolerance.
Appropriate in-house developed Software is available for all analyses, but has to be adapted.

Specific Objective 3: Association Mapping and Model Development

Target Output 3: Effects of specific CG alleles or CG allele combinations on the tolerance levels of the analysed stresses detected through Association Mapping and Models allowing predictions of parental breeding values and progeny performances established.			
Activity	**Project outputs**	**Targeted Outputs (Deliverable)**	**Due date**
A3.1 Analysis of potential associations of specific marker alleles with specific characteristics	Results of potential associations of specific marker alleles or combinations with specific characteristics such as tolerance levels and production	**D3.1a,b:**LIST of effects of alleles and AC of each CG on stress tolerance levels and production.	Months* 24 and 32
A3.2 Allele specific primers	Results of the PCR assays of allele specific primers for important CG alleles	**D3.2a,b:** LIST of allele specific primer sequences and their applications	Months* 24 and 32
A3.3 Initial Model Development	Identification of powerful models to predict progeny performances with respect to tolerance to abiotic/biotic stresses and production.	**D3.3a,b:** Initial ESTIMATES of Parental Breeding values and Progeny performances. LIST of most promising recommended crosses for the 3rd year.	Months* 24 and 32
A3.4 Model Validation and Refinement	Results of Model Validation and Model Refienment	**D3.4a,b:** Model PARAMETERS of optimized models. LIST of most promising crosses recommend for the future, based on these results.	Month 36

Activities:
A3.1: To perform Association Mapping for detecting associations of specific marker alleles with levels of stress tolerance in the set of 210 accessions from the field trials (A1).
A3.2: To design allele specific primers (ASP) for important CG alleles
A3.3: To develop models for marker assisted selection (MAS) by assigning parental breeding values and progeny performance predictions.
A3.4: To validate and refine these models based on observed progeny performances in Activity 4.2

Methodology

Activity 3.1- Association Mapping to detect CG allele effects

Based on the results from Activities 2.3 and 2.4, Partner **P4** will analyse the potential effects of specific candidate gene alleles (or combinations of such alleles) on the phenotypic expression of the corresponding trait. For these purposes association mapping techniques based on LDA ("linkage disequilibrium analysis"; Luo et al. 2000) will be applied according to Yu et al. 2006 and Abdurakhmonov & Abdukarimov 2008, and particularly using the mixed-model approach (Stich et al. 2008).

Activity 3.2 Design of allele specific primers (ASP) for important CG alleles

Based on observed sequence differences between CG alleles it is possible to design primers, which amplify selectively only specific alleles. ASP development is complex and multiplexing is difficult due to varying, specific PCR conditions. Therefore, Multiplexed PCR and Amplicon sequencing will be the method of choice with dropping sequencing costs when analysing a larger number of CGs.

Nevertheless, Allele-specific primers will be designed for single alleles of selected important genes, which contribute very significantly to phenotypic trait expression. These can be used for rapid screening of individual CG alleles. Their functionality will be evaluated in test amplifications on a small subset of the original genotype set.

Activity 3.3 Initial Model Development

Based on the **results from Activity 3.1**, we will perform Model building for MAS considering multiple CG, These Models will allow to estimate parental Breeding Values (**BV)** and to predict Progeny performance (**PPP**).

We will establish **Allele Models** (AL) and **Allele Combination Models** (AC) using multifactorial analyses (**Proc GLM, Proc Mixed**) applying the following steps:

1. **Selection of specific CG for the Model**, based on significances of individual CG effects and CG value correlations.
1**. Assignment of values to alleles and AC** based on the performance of the genotypes (GT) where they appear.
3. **Assignment of Parental Breeding values** (**BV**; only for AL Models) based on the average value of the alleles of each parent, averaged over all selected CGs)

We will establish **PREDICTIONS of PROGENY PERFORMANCE**:

For **AL Models** = based on the Average Parental Breeding Value
For **AC Models** = Average AC value of expected AC depending on parental alleles, averaged over all selected CG
The best Model is supposed to have the highest correlation between predicted and observed values. In-house developed Software (**TAMAS**) is available for these analyses and will be adapted.
Based on these predictions an initial List of recommended crosses will be established. This list will be used for performing the crosses in the 3rd year (Activity 4.2)

Activty 3.4 Model Validation and Refinement

A) Model Validation: Using the parameter estimates of **A3.3** for each CG and the molecular data (allele composition of GT) from **A2.3 and A2.4** it is also possible to make Progeny Performance Predictions for the progenies from Activity 4.2, below.
On the other hand NEW, observed progeny performance data for the traits of interest will be obtained in Activity 4.2. Thus, by comparing predicted model data (PV, PPP) and observed phenotypic data of the Progenies from Activity 4.2, it is possible to validate and proof the general applicability of a MAS Model, if significant correlations are obtained. Therefore, alternative MAS models will be validated based on correlation analyses, by paired T-Tests or Wilcoxon signed-rank tests using SAS Software (SAS 1989). The BEST MAS model can be determined in this way.
B) Model Refinement: The phenotypic data from Activity 4.2 will allow establishing new models as described in Activity 3.3, but here based on the expected allele configurations in the progeny genotypes, and allow to estimate new PV and PPP values. By combining the results of the initial validated models and these new models, it will be possible to refine the existing Models for optimal predictions.

Specific Objective 4: Pre-breeding activities to combine favourable characteristics, to improve adaptation to climate change, and to improve progeny performance predictions

Target Output 4: Genotypes with combined favourable characteristics obtained through pre-breeding activities and application of developed markers, allowing to improve adaptation to climate change and progeny performance predictions.

Activities	Project outputs	Targeted Outputs (Deliverable)	Due date
A4.1. Crossings	Crossings between promising accessions in order to combine favourable characteristics	**D4.1a,b,c:** List of performed crossings and parents involved	Months* 12, 24 and 36
A4.2. Evaluation of resulting progenies	Results of progeny evaluation and Selection of superior breeding clones with combined favourable characteristics.	**D4.2a,b:** Data of Progeny evaluations, LIST of selected progeny genotypes	Months* 24 and 36
A4.3. Application of MAS	Results of application of the developed allele specific primers in selected progeny genotypes.	**D4.3a,b:** Data of marker validation in selected genotypes	Months* 24 and 36

Activities:
A4.1: To perform crosses between accessions from the field trials (A1)
A4.2: To evaluate the resulting progenies in the field.
A4.3: To apply the developed allele-specific primers for progeny genotype selection.

Methodology:

Activity 4.1 Performance of crosses between accessions from A1
Partners **P1**, **P2** and **P3** will perform each year crosses using the accessions which are evaluated in WP1 as parents. The aim is to combine favorable characteristics of the parents or even superior CG alleles with respect to stress tolerance. For the crosses of the 3rd year, the recommendations provided by the initial Models (Activity 4.3) will be applied.

Activity 4.2 Evaluation of the obtained progenies for agronomic performance and tolerances

The obtained progenies will be sawn in the field at locations with adverse conditions and evaluated for their agronomic performance in order to select genotypes with superior characteristics. Participatory selection involving local farmers will be applied.

Activity 4.3 Application of the developed allele-specific primers for genotype selection.

At the same time the allele-specific primers which have been developed in Activity 3.2 will be applied to check the most promising genotypes in the progenies for the presence of favourable alleles.

- *Evaluation data will be also used for model validation and refinement as described in detail in Activity 3.3.*

Specific Objective 5: Dissemination and Transfer of Project results and Products

Target Output 5: Efficient Dissemination and Transfer actions realized to implement successfully Project results and Products.

Activities	Project outputs	Targeted Outputs (Deliverable)	Due date
A5.1. Knowledge Base	Establishment of a Knowledge Database about "Analysis and Evaluation of tolerance/ resistance to stresses in the potato crop.	**D5.1:** Knowledge Database based on all project results and external information	Month 6 and updates month 12, 24,36
A5.2. Project WEB page	Establishment of an informative Project web page	**D5.2:** Project WEB Page "Papaclima" with regular updates	Month 6 and updates month 12, 24,36
A5.3. Scientific Dissemination	Efficient transfer of project results between project partners and to the scientific community	**D5.3:** Publications of project results. Presentation of project results in conferences and workshops.	Periodically during the project
A5.4. Transfer of results to the sector	Efficient transfer actions to the sector (productive cahin) and stakeholders	**D5.4:** Fairs, Field Days, Regional Workshops. Distribution of tubers	Periodically during the project
A5.5. Demonstration Plots	To establish Demonstration plot s with most promising varieties or breeding clones	**D5.5:** Demonstration Plots of adapted varieties /breeding clones	Months 30 to 36

Activities:
A5.1: To establish a Knowledge Base on Tolerance/ Resistance to stresses in Potato
A5.2: To establish a Project WEB page
A5.3: To disseminate project results through publications and congress presentations
A5.4: To transfer project results and products between partners and to the potato productive chain
A5.5: To establish Demonstrative plots of most promising accessions or breeding clones.

Methodology

The consortium members consider several instruments and numerous actions to efficiently disseminate, transfer and exploit technology, knowledge, materials and other project results.

Activity 5.1: Establishment of a Knowledge Base on Analysis and Evaluation of Resistance / Tolerance to stresses in potato

All phenotypic and molecular data obtained in the project, the results of association mapping and model building including the applied methodology will be compiled into a Knowledge Database "Analysis and Evaluation of tolerance/ resistance to stresses in the potato crop." All partners will provide the necessary input.

Activity 5.2: Establishment of a Project WEB Page

Participant **P4** will establish the Project WEB page: "PAPACLIMA" with information about the project and its partners, along with all results, which are being obtained. The Knowledge Base will be part of this website. For this purpose all participants will send relevant information and results to P4.

Activity 5.3: Dissemination at the scientific / technical level

Scientific and informative articles, contributions to conferences and training courses will be realized. For internal transfer between partners, the annual meetings will be combined with technology transfer courses. The technologies will include phenotypic and agronomic assessments, specific molecular techniques, bioinformatics and statistical methods.

Activity 5.4: Transfer to the sector (productive chain)

Fairs and Field Days with farmers, breeders, experts and other actors of the potato productive chain (stakeholders) will be organized to inform about the project and project results, to show the field trials, to present adapted varieties and to distribute tubers of recommended varieties for cultivation.

Regional workshops will be held with farmers and farmer associations to present and discuss new knowledge and practices. Recommendations for growing potatoes and proper handling will be given.

Activity 5.5: Establishment of Demonstration Plots

Demonstration fields will be established in harsh environments in order to show adapted varieties / breeding clones with a good agronomic performance under these conditions. Thus, farmers and experts can check the value of these varieties for sustainable agriculture.

2.4. Targeted PGRFA

The project targets the potato crop in a broad sense and will include commercial *S. tuberosum* cultivars, local varieties or landraces, breeding clones, but also some native and wild potato species, which belong to other cultivated tuber-bearing Solanum species (non *S. tuberosum*).

The following plant material will be used in the project, or produced (= progenies) within the pre-breeding activities. In addition, the location of the field trials is indicated and their characteristics (**Table 1**). **Table 1.**

Partner	**Nº of Accessions***	**Field Trials for Evaluation**			**Pre-Breeding**
		Location	**No of Acc.**	**Stress**	**Nº of crosses/ Progenies**
P1 IBT	1.= 25 2. =50 3. =20 4. = 10	Fields Huancavelica Fields Quilcas	60 60	Drought /Control Cold	120/60
P2 INIAP	1. =30 2. =30 3. =30 4. =20	Sta. Catalina Greenhouse Field at Sta. Catalina Field at Chimborazo	50 50 70	Heat/Drought Control/Drought Cold	100/50
P3 ULA	1. =30 2. =40 3. =20 4. =20	Field at Merida Field at Trujillo	60 60	Control/Drought Cold	120/60

1. Commercial Cultivars, 2. Local Landraces, 3. Breeding clones, 4. Accessions of native or wild Potato species

2.5. Target groups and beneficiaries

The Andean farmers will have already after one year of project execution recommended cultivars or accessions at their disposal which can be grown under harsh and adverse agro-climatical conditions. In the near future through the pre-breeding activities in this project, potato varieties with improved properties such as tolerances and resistances to abiotic and associated biotic stresses which are adapted to the global climate change are obtained for sustainable agriculture. Supported by the different planned dissemination actions we expect to target over 500 farmers and their families in each country.
The knowledge and molecular data generated by the project are useful to increase the information about the entries of a Germplasm Bank, and can be integrated in the passport data. They provide guidelines for the functional biodiversity conservation of useful gene alleles for characters of interest, improving the representativeness and usefulness of the entries in a Germplasm Bank. The available information increases the use of such banks by the breeders, due to the availability of markers for assisted selection in genetic improvement programs.
Potato breeders will have improved breeding clones as progenitors at their disposal which can be used to develop novel potato varieties. We will address at least 20 Curators of Germplasm Banks and 30 potato breeders.
Breeders and researchers will have a set of molecular markers and predictive models useful for assessing adaptation to abiotic stresses in germplasm, progenitors and breeding clones at their disposal, which can be used to develop novel potato varieties.
The applied concept, using potato as a model species of the genus *Solanum*, can be potentially applied to other related species and crops.
The project WEB page and the foreseen congress presentations of project results will allow targeting at least 2000 scientists.

2.6. Impact and impact pathways

2.6.1. Food security and poverty alleviation

Our project will identify in a short-term varieties with better adaptation to adverse environmental conditions which are suitable for cultivation in disadvantaged zones. Thus, our project will contribute to the adaptation of the potato crop to the possible threats posed by climate change and prevent significant losses in production. These threats are closely related heat, cold and water availability and increased incidences of pests and diseases.
The cultivation of suitable genotypes will increase the income of farmers, thus contributing to sustainable development, food security and sovereignty and increase the quality of life and peace in this region.
The project will develop in a medium-term through the foreseen breeding activities also new potato cultivars with even higher tolerance levels to the analysed stresses, by combining favourable characteristics of the progenitors. Thus, at the end of the project farmers will have improved potato varieties adapted to extreme climatic conditions for sustainable agriculture at their disposal. This will lead to additional income, improved life standards and increased financial capacities for new investments or additional purchase of consumables.
The numerous foreseen dissemination and transfer actions to farmers and all actors of the potato chain will ensure the efficient implementation of all project results and products.

2.6.2. Adaptation to climate change and environmental sustainability

The availability of suitable varieties for adverse environmental conditions will improve the competitiveness of the potato crop, increase the area of cultivation and diversify agricultural production.
Through the provision of varieties tolerant to extreme environments, the project aims to contribute to the Millennium Development Goals (1, 7 and 8). It will enable to expand the agricultural frontiers for potato cultivation and will favour inhabitants in regions with extreme climates, allowing them access to new sources of nutrition and income. At the same time this

will allow to cultivate areas that previously did not have alternative crops, reducing in this way the impact of desertification.

2.6.3. Scientific impact

The knowledge and materials generated by this project will accelerate significantly potato breeding programmes to obtain improved varieties for sustainable agriculture adapted to climate change.

The specific candidate gene alleles with low and high values for adaptation to climate change which will be detected, provide guidelines for functional germplasm conservation to gene bank curators which should aim to particularly conserve the most useful CG alleles. Valuable accessions will be maintained in this way and can be exploited by potato breeders.

However, the predictive models developed in this project will have a very practical impact, far beyond the application of several individual markers for MAS, by suggesting exactly the most promising crosses which should be performed for most efficient breeding.

2.6.4. Capacity development and empowerment

The foreseen transfer of all generated project results, knowledge, methodologies and Software will strengthen the capacities of the partners in this project, which in turn will transfer the acquired items to their national researchers and project target groups.

After project completion they will be able to launch analogous projects in other related crops or other research topics using the applied strategy methodologies of this project.

2.7. Relevance to national or regional priorities in its plans and programmes for PGRFA

The governments of the Andean countries have made tremendous efforts to protect and use biodiversity in a way that can ensure its use without adversely affecting the natural habitats. Bio-prospecting plays an important role in conservation and utilization of these resources. For potato, the characterization and safeguard of the local landraces, wild and native species growing in the Andes will allow its use for the potato farmers and will reduce the environmental impact of the excessive use of agrochemicals.

The governments of Peru, Ecuador and Venezuela have launched National Food Plans, which gives special importance to the cultivation of the potato. It is therefore strategic for the countries, to count on new varieties adapted to the increasing threats related to climate change and its ecological and economic implications. At the same time, viable alternatives for the production and export of potato to other countries have emerged, either as seed or for fresh consumption. The participating institutions have taken a leading role in the new government strategies and therefore on the breeding and cultivation of potato.

For NEIKER the interest lies in the knowledge about candidate genes for climate change and molecular markers which will be generated through this project. These can be transferred to or exploited in other genetic backgrounds and even related crops.

SECTION C: OPERATIONS

3.1. Methodology of project implementation

Right at the beginning of the project a Consortium Agreement will be signed between partners in order to establish the legal and technical aspects of the collaboration.

We will apply up-to-date Methodology to implement the Project (see: The Basics of Project Implementation, CARE US; http://www.careclimatechange.org/files/toolkit/CARE_Project_Implementation.pdf).

The components of a successful project include managing relationships between partners and with various stakeholders, managing human resources, managing financial resources, facilitating learning, managing risks and ensuring flexibility.

Some of the elements for project implementation are already included in this proposal, such as objectives (2.2) a detailed work plan (2.3) including activities and executing partners, expected outputs and time schedule of activities, project budget (Appendix IV) and a logical framework (log frame) that explains how the project will contribute to the ultimate impact (Appendix II). Others will be implemented at project start, such as a Monitoring and Evaluation Plan, Budget Planning and Monitoring including a Staffing & Procurement Plan. Moreover, an Annual Work Plan (AWP) will be prepared, containing a detailed planning of the foreseen activities and deliverables and the specific set of results to achieve during a particular year. In order to control and monitor the progress of the project the following elements will be implemented:

Project Management Board (PMB)

The PMB will monitor the progress of the project. It is composed of the Coordinator and the principal investigators of each participating institution, together with the corresponding heads of departments of each institution as external observers and project staff, if required.

The coordinator in collaboration with the other PMB members will be responsible for reporting and the technical and financial management of the project.

Communication flow and Progress Control

General communication between the project partners will be realized via email or Skype. At each anticipated milestone the responsible partner(s) will write a brief report, stating whether the milestone has been met and, if not, the reason and a new expected date when the milestone will be met. In addition, to enhance the exchange of data and ideas and to enable the coordinator to closely follow the progress (and interfere if necessary) it is expected that each partner regularly provides the coordinator with informal progress reports, who will take care of the distribution of the information via email.

In order to facilitate the monitoring of activities and progress, a chronogram for the planned R&D tasks has been established (Appendix III).

Reporting in appropriate format will be performed as requested by the financing agency.

Annual meetings will be realised to plan and coordinate the R&D activities, to present and discuss the obtained results and to realize TT courses.

Management of knowledge and intellectual property

The project generates new knowledge in different fields as described above. Several instruments are implemented to disseminate freely this knowledge and associated technologies and results. These include beside publications also congress contributions and a detailed Project WEB page. No restrictions exist upon the diffusion of results and germplasm.

3.2. Partnerships and collaboration arrangements

The Leader of this research project is the National Agrarian University La Molina (UNALM, Lima, PERU) and particularly the Institute of Biotechnology (IBT) of UNALM **(P1).** IBT is at the forefront of scientific research in Peru and started its activities in 1998 with the participation of researchers and professors from different UNALM faculties (Agronomy, Biology, Forestry, Animal Science and Food Industries). One of the three main research areas at IBT focuses on the economic potential of native species (including Andean root and tuber crops). UNALM collaborates in this project with the co-participants, Universidad de los Andes (ULA, Mérida,

Venezuela), and the National Agriculture Research Institute – INIAP (Ecuador). The National Institute for Agrarian Innovation (INIA, Peru) will also be involved. There will be collaborative research and capacity building roles by the International Potato Center, Lima.

The main activities of INIAP include agricultural R&D and the application of scientific knowledge and technological innovations focusing on the rational exploitation and preservation of natural resources including Andean tuber crops.
ULA belongs to the superior education network of Venezuela. The Faculty of Sciences is the most productive faculty in the ULA, and hosts the Instituto Jardín Botánico de Mérida which houses the molecular biodiversity and variability lab (MBVL). The MBVL has previous experience in field research and laboratory research concerning the use of molecular techniques to identify and characterize germplasm resources of different species.

All three partners collaborate in this project with NEIKER (Basque Institute for Research and Development in Agriculture; Vitoria-Gasteiz, Spain) as sub-contractor for Technology Transfer. In years 2 and 3 there will be capacity strengthening mid-stays at Neiker for the project personnel of each participating country. NEIKER has realized different genomic studies in a dozen of plant species applying different molecular tools such as transcriptome mapping, differential expression analyses, microarray analyses or NGS sequencing technologies. Actually NEIKER participates in large Genomic Projects in oil palm and *Acacia*. The strategy and methodologies (including Software) which will be used in this project have been already applied successfully in these crops.

3.3. Project management team

The composition of the project management team is given below for each collaborating institution:

P1. UNALM-IBT (Peru):

Prof. Dr. Enrique N. Fernández-Northcote has been a Professor at the National Agrarian University for 25 years. He was formerly on the scientific team at the International Potato Center and became a Biosafety Consultant in 2002. He has significant experience in the field of Plant Pathology, specifically the genetic and molecular basis of resistance to plant diseases, and the evaluation and sustainable use of genetic resources combining biodiversity and modern biotechnology. He is currently a Visiting Professor at UNALM and Consultant on Modern Biotechnology and Biosafety at the IBT-UNALM. He was the National Coordinator for Peru in the LAC-Biosafety Project. As coordinator, he will be responsible for the overall technical and financial project management, the performance and evaluation of the field trials and for the dissemination and promotional events.

Dr. Raúl Blas is a Principal Professor at UNALM (Crop Husbandry Department, Agronomy Faculty) and a scientist at the IBT-UNALM with more than 15 years' experience particularly in genetic resource management, molecular biology and genetics. He will be responsible for the breeding activities and molecular analysis.

The National Institute for Agrarian Innovation (INIA), N.N. Master Students, Technicians and field workers will collaborate in molecular analysis, field trials and phenotypic evaluations.

P2. INIAP (Ecuador):

Dr. Xavier Cuesta scientist in charge, has 17 years' experience in breeding for resistance and quality traits in potato. He is responsible for the Integrated Potato Crop Technology area in the Potato Department. He will be responsible for the technical and financial project management at INIAP and for dissemination actions and promotional events.

MSc. Jorge Rivadeneira has 13 years' experience in breeding for resistance to late blight in potato, and is responsible for the potato breeding area. He will establish the germplasm collections and perform field trials and breeding activities.

MSc. Cecilia Monteros has more than 20 years' experience in Andean crop research including potato landraces. She will be responsible for the phenotypic evaluations and molecular analyses.

N.N. Technicians, Field workers and **Master Students** will collaborate in all project tasks.

P3. ULA (Venezuela) :

Dr. Gustavo A. Fermin M has been coordinating the MBVL and teaching the undergraduate courses in Genetics, Genetic Engineering and Plant Morphogenesis at the same faculty. He has co-authored several book chapters on economically-important viruses, as well as many scientific articles in the fields of plant pathology, plant biotechnology and plant biodiversity and variability. He will be responsible for the technical and financial project management at ULA, the organization of field trials and the dissemination and promotional events.

MSc. Carla Aranguren is a biologist with a Master in Ecology (specifically plant ecophysiology) with large experience in crop breeding. She will be responsible for the phenotypic evaluations, the breeding activities and the molecular analyses.

N.N. Master Students, Technicians and **Field workers** will collaborate in all project tasks.

P4: NEIKER

Dr. E. Ritter has over 20 years' experience particularly in breeding, molecular biology and genetics, and has participated in and managed many international projects. He will be responsible for the technical and financial management, the statistical analyses and the technology transfer courses.

Dr. JI Ruiz de G. is researcher at NEIKER and has over 15 years' experience in potato breeding. He will be responsible for the bioassays and in silco mining techniques and collaborate in the statistical analyses.

Dr. L. Barandalla has 12 years' experience in molecular marker technology as part of many previous R&D projects. She will be responsible for the generation of molecular data (molecular analysis).

NN additional **researchers** of the scientific team and **technicians** with relevant experience will collaborate in specific project tasks.

3.4. Sustainability

All partners have the necessary land and lab facilities as well as adequate financial, human and institutional capacities to perform successful the planned R&D activities in a sustainable way. Moreover, they know each other well and have already collaborated in the frame of other international R&D projects. They are also well connected to the government and all actors of the potato chain, ensuring in this way an efficient implementation of the project results and products. The project will allow the interaction of researchers from developing and developed countries, which will be of great benefit to both parties. This would settle the basis for the effective dissemination of results and future joint projects. It also gives an important benefit to native and wild species of potato which are hitherto unknown, and that greatly enhance the development of new potato varieties with tolerance to the major abiotic stress factors. Additionally, this research project will stimulate further collaborations between project partners on the same or related topics. Candidate genes and markers can be exploited in related (wild) species (tomato, pepper, aubergine) and perhaps in more distant species.

Availability of knowledge, materials and markers will improve considerably the competitiveness of the collaborating institutions as partners for such further R&D projects.

SECTION D: ANNEXES AND APPENDIXES

Annex 1

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Annex 2

ACTIVITY RESPONSABILITIES BY PARTNERS

Activity **A1**		**Start:** Month 1		**End:** Month 24
Participants:	P1: IBT	P2: INIAP	P3: ULA	
Man-months:	25 PM	30 PM	30 PM	• *PM=Person Month*
Activities/Deliverables**:**	**A1.1, A1.2** / D1.1ab, D1.2ab			

Activity **A2**		**Start:** Month 1		**End:** Month 24	
Participants:	P1: IBT	P2: INIAP	P3: ULA	P4: NEIKER	
Man-months:	6 PM	6 PM	6 PM	20 PM	
Activities /Deliverables:	A2.1, A2.2, A2.3, A2.4 / D2.1ab, D2.2ab, D2.3ab, D2.4ab				

Activity **A3**		**Start:** Month 20		**End:** Month 36	
Participants:	P1: IBT	P2: INIAP	P3: ULA	P4: NEIKER	
Man-months:	3 PM	3 PM	3 PM	10 PM	
Activities /Deliverables	A3.1, A3.2, A3.3 , A3.4 / D3.1ab, D3.2ab, D3.3ab, D3.4				

Activity **A4**		**Start:** Month 12		**End:** Month 36	
Participants:	P1: IBT	P2: INIAP	P3: ULA		
Man-months:	19 PM	21 PM	21 PM		
Activities /Deliverables	A4.1, A4.2, A4.3 / D4.1ab, D4.2ab, D4.3a,b				

Activity **A5**		**Start:** Month 1		**End:** Month 36	
Participants:	P1: IBT	P2: INIAP	P3: ULA	P4: NEIKER	
Man-months:	10 PM	9 PM	9 PM	4 PM	
Activities /Deliverables**:**	A5.1, A5.2, A5.3, A5.4, A5.5/D5.1, D5.2, D5.3, D5.4, D5.5				

APPENDIX 1: INFORMATION ON THE APPLICANT

Lead Organization (P1): **UNIVERSIDAD NACIONAL AGRARIA LA MOLINA (UNALM) – Instituto de Biotecnología (IBT)**

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APPENDIX 2: LOGICAL FRAMEWORK

ct title: MARKER ASSISTED SELECTION FOR POTATO GERMPLASM ADAPTED TO BIOTIC AND A STRESSES CAUSED BY GLOBAL CLIMATE CHANGE

	Intervention logic	Indicators/targets	Sources and means of verification	Assum
t	To contribute to the achievement of Millennium Development Goals 1 and 7: • *To eradicate extreme poverty and hunger* • *Ensure environmental sustainability*	1) At least 1500 farmers and their families benefit from the use of recommended or developed cultivars with adaptation to the threads of climate change which avoid production losses ensuring	- National Agricultural Statistics - Out site-	- Absence extreme w conditions catastroph - Lack of

		in this way food security and increased income of local farmers. 2) Potato cultivation increases at least 10%, particularly in regions with adverse agro-climatical conditions, extending the frontiers of potato cultivation and reducing in this way the impact of loss of potato diversity or desertification.	specific studies and surveys of the participating entities	order prob the region project ex - Absence unforesee changes i productio systems a relationsh agricultur output pri - Absence unforesee significan changes i prices of inputs (va rate of the
me	To improve adaptation and resilience to climate change and enhance the food security of resource-poor farmers in selected developing countries, by strengthening the sustainable management of plant genetic resources for food and agriculture (PGRFA).	Details of the concrete outcome are given in the different concrete outputs described below	Project outputs (see below)	- Farmers, breeders, actors of t potato cha gene bank are interes the projec outcome

t 1: Andean o varieties ccessions ling Native o species esistance or nce to c and ated biotic es related to l climate e identified, mended for ation under se conditions sed for ation and ing.	To carry out field trials and bio assays to evaluate agronomic performance and tolerance to abiotic stress factors: drought, cold, heat and resistance to associated biotic stresses (*P. infestans*). To identify promising, adapted or resistant accessions.	1.1) Over 300 PGRFA listed in *Annex 1* of the Treaty (Potato, *Solanum* spp. except *S. phureja*) made available according to the terms and conditions of the Multilateral System 1.2) Over 300 varieties, local landraces, breeding clones and other accessions documented, analysed and phenotyped for abiotic and associated abiotic stress tolerance 1.3) Over 30 promising accessions with elevated tolerance levels to abiotic stresses and associated biotic stresses identified and recommended for cultivation 1.4) Over 30 promising accessions with elevated stress tolerance levels identified and recommended for breeding.	**In the Deliverables:** **D1.1a,b:** Evaluation Data & Recommended LIST of accessions with tolerance to different abiotic stresses for cultivation & breeding **D1.2a,b:** Evaluation Data & Recommended LIST of accessions with resistance to *P. infestans* for cultivation & breeding - Annual Project Reports with justified Deliverables - Scientific and informative publications - Congress Proceedings - Press Notes - Project WEB page with all results -	- Presence of sufficient natural phenotypic variation with respect to stress response in the selected plant materials of the partners. .

			Documentation of Dissemination and Transfer events. - Documentation of Demonstration plots	

Output 2: Useful candidate genes for abiotic stress and associated biotic stress tolerance identified applying RNAseq, in silico Mining and RAD sequencing & existing allelic variation for these CG in the evaluated accessions determined.	To detect useful CG for adaptation to abiotic and associated biotic stress tolerance in potato applying RNA-Seq, analyzing published known genes from potato and other species and by RAD sequencing and CG extraction. To analyze the allelic variation in these Candidate genes and the CG allele composition in the accessions.	2.1) At least 200 useful CG for tolerance to the different analysed stresses with relevant biological meaning detected and exploited for the development of improved, stress adapted cultivars. 2.2) At least 400 useful alleles discovered in these CG and exploited for the development of new, stress adapted potato varieties	**In the Deliver** **D2.1a,b:** List c candidate gene abiotic and rela biotic stress res derived from R **D2.2a,b:** List c CG derived fro silico mining o published cand genes for abiot related biotic s resistance. **D2.3a,b:** List c Sequences and functional prim obtaining CG amplicons. For each CG L SNP/alleles in collection and composition of accession. **D2.4a,b:** LIST extracted from tags and their b meaning. LIST SNP/alleles in collection and composition of accession.

Output 3: Effects of specific CG alleles or CG allele combinations on the tolerance to the analysed stresses detected through Association Mapping and Models allowing predictions of parental breeding values and progeny performances established.	To perform Association Mapping for detecting effects of specific marker alleles on stress tolerance levels in the evaluated accessions, and to design allele specific primers (ASP) for most important CG alleles with large effects. To develop models for marker assisted selection (MAS) by assigning parental breeding values and progeny performance predictions and to validate and refine these initial models based on observed progeny performances.	3.1) At least 100 significant effects of specific marker alleles on stress tolerance levels detected. 3.2) At least 30 allele specific primer pair for most important CG alleles validated a provided. 3.3) At least 10 novel technologies relate marker assisted selection systems, metho and techniques for genetic improvement a conservation, bioinformatics, etc. co-developed and transferred. 3.4) Different models (AL or AC models individual and combined stresses) of mar assisted selection systems introduced and disseminated. 3.5) Three specialized bioinformatics too (ASPAM, RADSAT, TAMAS) made available, transferred and deployed for integrated data analysis and interpretation germplasm, genomic and phenotypic data

Output 4: Genotypes with combined favourable characteristics obtained through pre-breeding activities and application of developed markers, allowing to improve adaptation to climate change and progeny performance predictions.	To perform crosses between accessions from the field trials in order to combine favorable characteristics, to evaluate the resulting progenies in the field, and to select improved progeny genotypes supported by the use of the developed molecular markers (ASP).	4.1) Over 30 identified, promising accessions with elevated stress tolerance levels used for breeding involving over 3(crosses. 4.2) Over 100 useful breeding population developed. 4.3) At least 9 new improved varieties wi elevated stress tolerance levels developed through participatory breeding methods. 4.4) Allele specific primers for MAS in a least 150 progeny genotypes applied.

Output 5: Efficient Dissemination and Transfer actions realized to implement successfully Project results and Products.	To establish a Project WEB page with an integrated Knowledge Base on Resistance/Tolerance to stresses in Potato with periodical updates. To disseminate project results through publications and congress presentations and to transfer project results and products between partners and to the potato productive chain, including the establishment of Demonstrative plots	5.1) Informative Project WEB Page "Papaclima" with integrated Knowledge database established and regular updates 5.2) At least 30 publications of project results and presentation of project results in conferences and workshops. 5.3) At least 18 public dissemination and transfer actions realized (Fairs, Field Days, Regional Workshops) including the Distribution o tubers from recommended cultivars and breeding clones. 5. 4) At least 6 Demonstration Plots of adapted varieties or breeding clones. 5.5) Over 20 of the identified promising accessions with elevated stress tolerance levels used for cultivation by farmers 5.6) Over 30 of the identified promising accessions with elevated stre tolerance levels used for breeding by other breeders. 5.7) At least 9 seed production and dissemination initiatives established, and 60 smaller units of planting material multiplied from the field trials and distributed. 5.8) "Passport" information of 200 accessions and associated genomic/phenotypic information systematized and disseminated. 5.9) At least 9 PGRFA institutions in developing countries benefiting from improved access to technologies and knowledge associated to adapted genetic material; 5.10) At least 300 resource-poor farmers trained and involved in the development of new varieties and other relevant technologies for climate change adaptation and strengthening food security. 5.11) At least 90 links with rural communities facing environmental changes strengthened. 5.12) At least 9 capacity development activities (e.g. training workshops, knowledge exchange sessions, etc.) organized. 5.13) At least 20 links established with national, regional and international gene banks.

		5.14) At least 30 links forged with research and development institutions regionally and globally. 5.15) The capacity of at least 15 local and national institutions strengthened to conserve, manage, improve and disseminate plant genetic resources. 5.16) The capacity of at least 18 lead developing country institutions, 1500 scientists and 300 stakeholders strengthened in the use of marke assisted selection systems.

APPENDIX 3: WORK PLAN (Gantt Chart)

Project title: **MARKER ASSISTED SELECTION FOR POTATO GERMPLASM ADAPTED TO BIOTIC AND ABIOTIC STRESSES CAUSED BY GLOBAL CLIMATE CHANGE**

		1st Year						2nd Year							
		Months						Months							
	Activity as Tasks	**2**	**4**	**6**	**8**	**10**	**12**	**2**	**4**	**6**	**8**	**10**	**12**	**2**	**4**
	Output 1: Andean potato varieties and accessions including Native potato species with resistance or tolerance to abiotic a stresses related to global climate change identified, recommended for cultivation under adverse conditions and used for cultiv														
	Task 1.1: Evaluation of resistance or tolerance to abiotic stress factors: drought, cold, heat	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**		
	Task 1.2: Evaluation of resistance or tolerance to late blight (Phytophthora infestans)				**X**	**X**	**X**				**X**	**X**	**X**		
	Output 2: Useful candidate genes for abiotic stress and associated biotic stress tolerance identified applying Mining and RAD sequencing & existing allelic variation for these CG in the evaluated accessions determined.														
	Task 2.1 Library construction and RNA-Seq			**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**		
	Task 2.2 Analysis of known candidate genes for biotic and abiotic stresses.	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**		
	Task 2.3: Analyses of CG by Amplicon Sequencing (CG driven approach)							**X**	**X**	**X**	**X**	**X**	**X**		
	Task 2.4: RAD sequencing for CG detection and analyses (random approach).							**X**	**X**	**X**	**X**	**X**	**X**		
	Output 3: Effects of specific CG alleles or CG allele combinations on the tolerance to the analysed stresses detected t Mapping and Models allowing predictions of parental breeding values and progeny performances established.														
	Task 3.1- Association Mapping											**X**	**X**	**X**	**X**
	Task 3.2 Design of allele specific primers (ASP) for important CG alleles														**X**
	Task 3.3 Model Development												**X**	**X**	**X**
	Task 3.4 Model Validation and Refinement														

		1st Year						2nd Year							
		Months						Months							
	Activity	**2**	**4**	**6**	**8**	**10**	**12**	**2**	**4**	**6**	**8**	**10**	**12**	**2**	**4**
	Output 4: Genotypes with combined favourable characteristics obtained through pre-breeding activities and application of allowing to improve adaptation to climate change and progeny performance predictions.														
	Task4.1 Performance of crosses between accessions from A1					**X**	**X**					**X**	**X**		
	Task 4.2 Evaluation of the obtained progenies for agronomic performance and tolerances							**X**	**X**	**X**	**X**	**X**	**X**	**X**	**X**
	Task 4.3 Application of the developed allele-specific primers for genotype selection.											**X**	**X**		
	Output 5: Efficient Dissemination and Transfer actions realized to implement successfully Project results and Products.														
	Task 5.1 Establishment of a Knowledge Base on Analysis and Evaluation of Resistance / Tolerance to stresses in potato		**X**		**X**		**X**		**X**		**X**		**X**		**X**
	Task 5.2: Establishment of a Project WEB Page				**X**		**X**			**X**			**X**		
	T5.3 Dissemination at the scientific / technical level		**X**		**X**		**X**		**X**		**X**		**X**		**X**
	Task 5.4 Transfer the sector (productive chain)			**X**		**X**		**X**		**X**		**X**		**X**	
	Task 5.5 Establishment of Demonstration Plots														

APPENDIX 4: BUDGET

See Budget attachment.

APPENDIX 5: DISBURSEMENT INFORMATION

Bank Name: BANCO DE CREDITO DEL PERU
Bank address: Esq. Juan de Arona y Rivera Navarrete s/n
Branch : SUCURSAL SAN ISIDRO
Country : PERU
Beneficiary : FUNDACION PARA EL DESARROLLO AGRARIO
Account number: 002-191-000417171158-55
Account currency: US Dollar
IBAN Code:
SWIFT Code: BCPLPEPL

APPENDIX 6: ENDORSEMENT LETTERS

See Letters attached from:

IBT-UNALM, LIMA-PERU

INIAP, QUITO-ECUADOR

ULA, MERIDA-VENEZUELA

NEIKER, VITORIA-SPAIN

By signing this submission form for full proposal, the applicant confirms that all the above statements, including the attached Appendixes, are true to the best of his/her knowledge. Any deliberately untruthful response will lead to the automatic exclusion from the further screening and appraisal process, and may lead to the denial of awarded grants from the Benefit-sharing Fund.

Signature of contact person:	**Date and location**
Enrique N. Fernández-Northcote	**December 04, 2014. Lima-Peru**