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# Identification of insect pests and diseases of potato cultivated in family farming in the Sacaba municipality, Bolivia

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# IDENTIFICATION OF INSECT PESTS AND DISEASES OF POTATO CULTIVATED IN FAMILY FARMING IN THE SACABA MUNICIPALITY, BOLIVIA.

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TRAVAIL DE FIN D'ÉTUDES PRÉSENTÉ EN VUE DE L'OBTENTION DU DIPLÔME DE MASTER BIOINGÉNIEUR EN CHIMIE ET BIOINDUSTRIES

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PROMOTEURS: LUDIVINE LASSOIS, JORGE A. ROJAS-BELTRAN

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# Résumé

La Bolivie enregistre de faibles rendements pour la production de pomme de terre par rapport à d'autres régions du monde. Les bio-agresseurs associés à la pomme de terre sont un des facteurs responsables de la réduction des rendements de la production de la pomme de terre. L'objectif du présent travail est d'apporter les informations nécessaires pour développer des méthodes de lutte appropriées contre les ravageurs et les maladies de la pomme de terre. L'étude se déroule dans le district de Palca (municipalité de Sacaba, département de Cochabamba, Bolivie) situé dans la région Andine. Tout d'abord, une description du système de production comprenant les variétés cultivées, la provenance des semences, l'utilisation/destination de la production, le cycle de rotation, les périodes de semis, les périodes de récolte, les pratiques agricoles, l'utilisation de produits phytosanitaires, les risques, et les problèmes liés à la production de pommes de terre a été réalisée. Ensuite, pour l'identification et la quantification des bio-agresseurs (insectes ravageurs et maladies bactériennes, fongiques et virales) associés à la culture, le travail a été réalisé dans trois parcelles de pommes de terre (variétés Waych'a et Pinta Boca) du 19 mars au 9 avril 2022. Le comptage des insectes a été réalisé avec des pièges von Moerick, des pièges à puits et des observations visuelles des plantes et des tubercules. Au cours de cette période, dans les trois parcelles, les ravageurs Phthorimae operculella, les pucerons (Myzus persicae et Rhopalosiphoninus latysiphon) et un coléoptère de la famille des Curculionidae ont été identifiés. Pour les maladies, 75 plantes et 3 de leurs tubercules ont été échantillonnés dans chaque parcelle. Pour chaque maladie dans chaque parcelle, l'incidence et la sévérité ont été mesurées. Les maladies identifiées sont la septoriose (Septoria lycopersici var. malagutii), l'alternariose (Alternaria solani), le mildiou (Phytophthora infestans), le rhizoctone brun (Rhizoctonia solani), la gale poudreuse (Spongospora subterranea f. sp. subterranea), la gale verruqueuse (Synchytrium endobioticum), la cercosporiose (Cercospora spp.) et le virus Y de la pomme de terre (PVY).

## Mots clés

Pomme de terre - Région andine - Bolivie - Bio-agresseurs - Insectes ravageurs - Maladies - Système de production

# Abstract

Bolivia registers low potato production yields in comparison to other regions of the world. The bioaggressors associated with the potato are one of the factors responsible for the reduction of potato production yields. The work aims to provide the necessary information to develop appropriate integrated pest and disease management strategies for potato production. The study takes place alongside the farmers of the Palca district (Sacaba municipality, department of Cochabamba, Bolivia) in the Andean region. Firstly, a description of the potato production system comprising the potato varieties cultivated, the provenance of the seeds, the use/destination of the production, the rotation cycle, the sowing periods, the harvest periods, the agricultural practices, the use of phytosanitary products, the risks, and the problems associated with the potato production was realised. Secondly, for the identification and the quantification of bio-aggressors (insect pests, bacterial, fungal, and viral diseases) associated with the potato cultures, the work was carried out in three potato plots (Waych'a and Pinta Boca varieties) from the 19th of March to the 9th of April 2022. The counting of the insect pests was realised with von Moerick traps, pitfall traps, and visual observations of the plants and the tubers. During this period, in the three fields, the pests Phthorimae operculella, aphids (Myzus persicae and Rhopalosiphoninus latysiphon), and a beetle of the Curculionidae family were identified. For the diseases, 75 plants and 3 of their tubers were sampled in each plot. The disease identification was based on symptomatology. For each disease in each field, the incidence and severity were recorded. The diseases identified were Septoria leaf spot (Septoria lycopersici var. malagutii), the early blight (Alternaria solani), the late blight (Phytophthora infestans), the black scurf (Rhizoctonia solani), the powdery scab (Spongospora subterranea f. sp. subterranea), the potato wart (Synchytrium endobioticum), the Cercospora leaf blotch (Cercospora spp.), and the potato virus Y (PVY).

# Keywords

Potato -- Andean region -- Bolivia -- Bio-aggressors -- Insect pests -- Diseases -- Production system

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## 1. Introduction

In terms of world production, potatoes production is placed in fourth place after wheat, rice, and corn. This ranking positioned potatoes as a vital crop for world food security. In Bolivia, potatoes and Andean tubers are major crops for the food security of the country. Furthermore, the high Andean region is the centre of domestication and genetic diversity of potatoes and Andean tubers. More precisely, around Titicaca Lake, the most primitive agriculture for the cultivation of potatoes, and other tuberous plants has been practiced. Even though potato production has been going on for 8000 years in this region of the world, the yield production is low. The average yield of Bolivia is only around 5 to 6 tons/ha. In the Andean region, the cultivation of potatoes is mainly realised in family farming. In that matter, the Fontagro developed a project to improve production. The Fontagro is a mechanism of cooperation working in 15 countries of Latin America, the Caribbean countries, and Spain for agricultural research, development, and innovation.

A reason for these low yields is the use of poor-quality seed tubers. To address the issue, the principal goal of the Fontagro is to produce good-quality seed tubers with the project Root to Food. The Universidad Mayor de San Simón decided to work on this project to bring the necessary technology to offer good-quality seed tubers. The cleaning of the seed is realised through in vitro tissue culture. The seeds of good genetic and phytosanitary quality are expected to be continuously available to the farmers. The distribution to the farmers should be realised with the help of the Sacaba municipality which is part of the department of Cochabamba in Bolivia.

To complement this objective, further knowledge of other problems that are causing low yields in the potato production of the Sacaba municipality is important. In addition to the bad-quality tubers, the bio-aggressors of the potato are also responsible for the reduction of the yields. As it is well known nowadays, the excessive and inadequate use of phytosanitary products to control bio-aggressors is damaging to the environment and human health. Consequently, knowledge of the bio-aggressors present in the Sacaba municipality is important to develop proper strategies in integrated pest and disease management programs.

The implementation of both strategies by the farmers of the municipality, the use of good quality seed for sowing, and integrated agricultural practices by the farmers will hopefully reduce their use of phytosanitary products as well as increase their production yields.

# 2.1. The biology of the potato 2.1.1. Taxonomy

The potato is classified into the *Solanaceae* family, the *Solanum* genus, and more precisely into the tuber-bearing species of the petota section<sup>1,2</sup>. A widely used classification of the petota section was realised by Hawkes in 1990 where the cultivated potatoes were classified into seven species<sup>1</sup>. However, Spooner et al. (2014) reviewed the early classifications of the petota section and determined that there are only four cultivated potato species<sup>2</sup>.

Potatoes present different ploidy levels<sup>1–3</sup>. From the Hawkes classification: the diploids species are *Solanum stenotomum* Juz. & Bukasov, *Solanum phureja* Juz. & Bukasov, and *Solanum × ajanhuiri* Juz. & Bukasov; the triploids are *Solanum Juzepczukii* Bukasov, and *Solanum × chaucha* Juz. & Bukasov; the tetraploid is *Solanum tuberosum* subsp. *andigena* Juz. & Bukasov and *Solanum tuberosum* subsp. *tuberosum* L., the pentaploid is *Solanum curtilobum* Juz. & Bukasov<sup>1</sup>. While the Spooner et al. (2014) classification is: "(1) *S. tuberosum*, with two cultivar groups (the Andigenum group of upland Andean genotypes containing diploids, triploids, and tetraploids and the Chilotanum group of lowland tetraploid Chilean landraces), (2) *S. ajanhuiri* (diploid), (3) *S. juzepczukii* (triploid), and (4) *S. curtilobum* (pentaploid)"<sup>2</sup>.

Among these species, only *S. tuberosum* ssp. *tuberosum* is grown worldwide while the other species are mainly restricted to the Andean countries.

#### 2.1.2. Botanical characteristics and development cycle

The potato is a dicotyledon autogamous herbaceous plant<sup>4</sup>. The aboveground vegetative system is annual while the underground vegetative system is perennial.

The aerial system is composed of several stems<sup>5</sup>. The stems are angular to round in cross-section<sup>5</sup>. The leaves are compound with 3 to 5 pairs of leaflets and a terminal leaflet. The inflorescence is a biparous cyme with 8 to 10 flowers. The flowers are pentamers with various colours<sup>4</sup>. The fruit is a round berry containing 200 to 300 seeds<sup>4</sup>. The underground system is composed of the roots, the stolons, and the tubers<sup>5</sup>. The tubers are tuberised extremity of stolons that form the main storage organ of the potato plant. On the tubers are present dormant eye-buds underlined with an aborted leaf <sup>5</sup>.

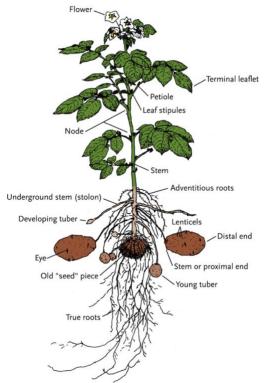


Figure 1: Morphological characteristics of a potato plant<sup>6</sup>.

The development cycle varies between 90 days for early maturing varieties of potato up to 150 days for late maturing varieties of potato<sup>3</sup>. The first stage of potato development regards the development of the sprouts from the mother tuber<sup>4</sup>. The sprout emergence from the ground leads to the vegetative growth stage<sup>4</sup>. During this stage, the sprouts of the aboveground system become the stems<sup>4</sup>. Leaves develop from the axillary buds of the stems<sup>4</sup>. In the soil, the stolons and the roots develop. Next, the ends of the stolons stop their development and start to tuberise<sup>4</sup>. In some cultivars, the tuberisation corresponds to the first flowering. The tubers will continue to grow until the senescence of the plant<sup>4</sup>.

#### 2.2. Potatoes production in Bolivia

#### 2.2.1. The situation of potatoes production in Bolivia

Bolivia produces more than 1.2 million tons of potatoes annually<sup>7</sup>. On average, 130 thousand hectares are allocated to potato production in the country<sup>4,8</sup>. Consequently, Bolivia's average production yield is 5 to 6 tons/ha whereas the worldwide average yield between 2002 and 2006 was 17.59 tons/ha<sup>8</sup>. In 2021, the department (dept.) of La Paz (Figure 18, cf. page 25) possessed the biggest cultivated area nationwide with 54,396 hectares and produced 247,419 tons of potatoes<sup>7</sup>. The dept. of Cochabamba was the second, with 42,453 hectares cultivated and 426,307 tons produced. In third place was the dept. of Potosí, with 30,045 hectares and 153,134 tons<sup>7</sup>. The other producing depts. are Oruro, and parts of Chuquisaca and Tarija<sup>7</sup>.

In Bolivia, each person consumes between 90 and 100 kilos of the tuber that is grown annually by 250 thousand Bolivian families<sup>8,9</sup>.

Initially, potatoes and Andean tubercules were only grown in the high Andes. Around the 1950s and 1960s, their production gradually spread to lower altitudes<sup>10</sup>. It started to spread in the inter-Andean valleys (2,000-3,000 meters-above-sea-level (masl)) and continued in the mesothermal valleys (1,000-2,000 masl) of the dept. of Santa Cruz, the Andean-Amazonian transition zone (1,000-2,000 masl), and the eastern plains (200-500 masl)<sup>10</sup>. Nowadays, potatoes and Andean tubers are cultivated from 800 masl up to 4,000 masl<sup>11</sup>. However, the production is still concentrated in the Andean region<sup>10</sup>.

In Bolivia, the Andean region is distributed among the depts. of La Paz, Chuquisaca, Potosí, Cochabamba, and part of the dept. of Oruro<sup>10</sup>. It covers three major areas<sup>10,12</sup>. First, there is the Altiplano (3,800-4,000 masl), which lies between the Western (*Azanaques*) and Central (*Los Frailes*) Andean ranges. The altiplano is a land of cold, high plains. Secondly, there is the *Puna* (3,000-4,500 masl), composed of rolling mountains with abutting slopes<sup>10,12</sup>. In the *Puna*, the climate can be cold and dry or cold and humid. Thirdly, there are the inter-Andean valleys (1,900-3,000 masl) and mesothermal valleys (900-1,900 masl), which lie between the mountain ranges and are irrigated by rainwater<sup>10,12</sup>. These areas present a climate that is temperate to temperate-hot and notably drier<sup>10,12</sup>. In the *Puna*, 73% of the total primary potato production is encountered. In the valleys, 20% of the total primary potato productivity of potatoes cultivation decreases with altitude while the diversity of potatoes varieties increases with altitudes<sup>8</sup>.

The Bolivian government, through the Institute of Agricultural and Forestry Innovation (INIAF), is implementing a series of policies to strengthen potato production in Bolivia. These policies promote the use of high sanitary quality potato seeds, fertilisers, crop protection agents, pressurized irrigations, etc<sup>10</sup>. Yet, the preservation of the genetic diversity of Bolivia's native potato varieties was overlooked<sup>10</sup>.

Indeed, in Bolivia, up to 1,095 varieties of the seven potato species are kept by the National Germplasm Bank of Andean Tubers and Roots<sup>4,13</sup>. This genetic diversity of potatoes and Andean tubers can be encountered in rural communities, especially in the depts. of La Paz, Cochabamba, and Potosi<sup>10</sup>. These rural communities permit the development of genetic diversity through their cultivation practices linked to their cultures alongside the climate and the geography of their localities<sup>10</sup>.

#### 2.2.2. Potatoes production system in Bolivia

Balderrama and Terceros (2009) identified five periods for sowing potatoes throughout the year. The right period to sow is a combination of the altitude, the environmental conditions, and the potato variety.

The *Lojru* sowing goes from February to March, the winter sowing from April to May, the *Mishka* sowing from June to July, the *Chaupi Mishka* sowing from August to September, and *Jatun Tarpuy* sowing from October to December. Over the period 2000-2006, the authors mention that 67% of the potato area is cultivated in the *Jatun Tarpuy*, 25% in the *Mishka* and *Chaupi Mishka*, and 8% in the *Lojru* and winter sowing<sup>11</sup>.

In Andean Bolivia, traditional technologies for potatoes cultivation are still used, especially where slopes are too steep<sup>10,14</sup>. For example, for the soil preparation, it includes *Yunta*, a Quechuan word for two yoked together, and manual tools such as the *Chakit'ajlla* for ploughing fields<sup>15</sup>. However, the use of agricultural machinery is also in application<sup>10</sup>. The agricultural machinery is used in some microcenters like the Altiplano around La Paz, the inter-Andean valleys, or high mountain areas where slopes permit<sup>10</sup>.

To continue, agrochemicals can be intensely used. In the traditional growing areas of Cochabamba and La Paz, fungicides are used to control late blight (*Phytophthora infestans*) and insecticides are needed to deal with thrips (*Frankliniella* spp.), aphids (*Myzus persicae*) and weevils (*Premnotrypes* spp. and *Rhygopsidius* spp.)<sup>10</sup>. Fungicides and insecticides are more used in wetter, colder areas<sup>10</sup>.

Whereas for fertilisation, the use of granular and liquid fertilizers is common, although chicken manure has been introduced to replace them<sup>10</sup>.

Finally, modern irrigation systems are rare with some exceptions in some microcenters where it is used during the winter sowing<sup>10</sup>.

# 2.3. The bio-aggressors associated with the potato crop in the Cochabamba dept.2.3.1. Insect pests present in the Cochabamba dept. in Bolivia

Various insect pests are responsible for damage to the potato culture in Bolivia. Figure 3 presents the stages of the major insect pests at the different physiological stages of the potato plant.

To start, there are Andean weevils which include several *Phyrdenus* spp., *Premnotrypes* spp. and *Rhigopsiudius piercei*, *Rhigopsidius tucumanus*<sup>4,16</sup>. The three weevils genera cause losses of 10% to 60%, up to 80%, and 25% to 85% for each weevil genera respectively<sup>4,16</sup>. *Phyrdenus* spp. is more prevalent in the mesothermal valleys from 1,500 to 2,700 masl and *Rhigospsidius* spp. at an altitude above 2,500 masl<sup>4</sup>. The weevil larvae feed on the tubers whereas the adults feed on the leaves starting at the margins leaving characteristic semi-moon shapes<sup>1,4,17</sup>.

*Phthorimaea operculella, Symmetrischema tangolias*, and *Paraschema detectendum* moths are also responsible for damages<sup>4,18</sup>. *P. operculella* and *S. tangolias* are respectively responsible for 50% to 100% and 30% to 80% of losses during storage<sup>4</sup>. Moreover, *S. tangolias* is more prevalent at higher altitudes (over 3,400 masl) compared to *P. operculella*. *P. detectendum* distribution varies between 2,740 and 4,000 masl, however, little information is available on this moth<sup>4,18</sup>. The damages are caused by the larvae of the three moths. The larvae feed on the tubers. To a minor extent, *P. operculella* causes damage to the leaves and *S. tangolias* to the leaves and stems<sup>4,17,18</sup>.

Flea beetle (*Epitrix* spp.) is present at altitudes between 2,000 and 3,500 masl<sup>4</sup>. The adults feed on the leaves leaving characteristic circular holes. Larvae feed on the underground organs and form characteristic galleries on the surface of tubers<sup>4,17,19</sup>.

Several aphid species are present in the potato crop such as *Myzus persicae*, *Macrosiphum euphorbiae*, *Aphis gossypii*, *Rhopalosiphum* spp.<sup>4,17</sup>. They are present in all the potato production areas of Bolivia but with a decreasing incidence over 3,000 masl<sup>4,20</sup>. They rarely cause direct damage by probing the plant sap but indirect damage by transmitting viruses<sup>4,17</sup>.

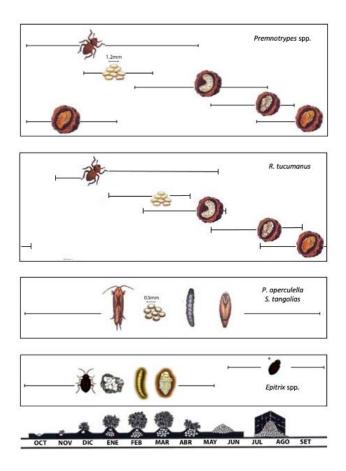


Figure 2: Stages of major insect pests at the different physiological stages of the potato plant for the Andean region of Peru (for *P. operculella*, *S. tangolias*, *Epitrix* spp.) and of Bolivia (for *Premnotrypes* spp., *R. tucumanus*)<sup>16,21</sup>.

Other occasional insect pests include white grubs, cutworms, thrips, and Epicauta spp.

White grubs of the *Scarabaeoideae* superfamily such as *Anomala incontestans* and *Phyllophaga* spp., were reported to cause damage from February to April in the Colomi municipality (Cochabamba dept.) from 3,000 to 3,350 masl<sup>4,22</sup>. The larvae damage the plant by feeding on roots, underground stem parts, stolons, and tubers of various plant species including potatoes<sup>4,17</sup>.

The cutworms are larvae of several noctuid moths such as *Agrotis* spp. and *Copitarsia* spp. They live in cold zones of the country between 1,500 and 3,700 masl. They are present in the Colomi municipality on November<sup>4,22</sup>. During the day, they remain in the soil at the base of the stems. However, during the night, they cut the stems and eat the leaves of young potato plants. They also damage tubers closer to the ground<sup>4,17</sup>.

Thrips (*Frankliniella* spp, *Thrips tabaci*, and other species) can cause severe damage in dry climatic conditions<sup>4,22</sup>. Thrips were identified as a risk in the Colomi municipality from November to February. Larvae and adults feed on leaves which develop a silver or chlorotic dotting<sup>4,17</sup>.

*Epicauta* spp. is present in all the potato production areas of Bolivia<sup>4</sup>. They are found in large groups attacking a plant until complete defoliation leaving only the stems of the plant<sup>4</sup>.

Finally, *Russelliana solanicola*, a psyllid present in the mesothermal valley of Cochabamba, and white flies (*Trialeurode vaporarium*) are additional pests of potato crop<sup>4,17</sup>. However, their preference for a more temperate to hot climate reduces the likeliness of their encounter<sup>4</sup>.

#### 2.3.2. Diseases present in the dept. of Cochabamba in Bolivia

A small number of diseases on the potato crop are induced by **bacteria** in the dept. of Cochabamba.

A first bacterial disease is the wilt or brown rot caused by *Ralstonia solanacearum*<sup>23</sup>. The pathogen was detected latently in some seed zones located above 2,800 masl<sup>4,24,25</sup>. It is a soil-borne bacterium that usually enters through wounds during the vegetative growing stage<sup>26</sup>. Wilting is a common symptom of the infection which starts at the youngest leaves (Figure 3B). The disease develops and becomes visible on one stem, one side of a plant, or the whole plant. The leaves may turn chlorotic or bronzed. In the stem, the xylem vessels become brown. When the stem is cut, a creamy mass exudes. Symptoms on the tubers are not always visible. However, if the symptoms are visible, the vascular ring of the tuber is necrotic and brown. As for the stem, when the tuber is cut, a creamy mass exudes (Figure 3A)<sup>4,17,27</sup>.



Figure 3: R. solanacearum tuber (A) and leaf symptoms (B)<sup>28</sup>.

A second bacterial disease is the blackleg or soft rot caused by *Pectobacterium. atrosepticum* and *Pectobacterium carotovorum* var. *carotovorum*<sup>4,29</sup>. The disease was identified in cold and humid localities at 3000-4000 masl and in localities of the inter-Andean valleys at 2000-3000 masl<sup>4,29</sup>. In these localities, the disease is responsible for little to moderate losses<sup>4,29</sup>. The major source of infections is latently infected mother tubers<sup>30</sup>. The mother tuber releases bacteria in the soil that will contaminate neighbouring tubers<sup>30</sup>. Daughter tubers can also be infected through the xylem<sup>30</sup>. The bacteria can enter the stems through wounds. Tuber and stem wounds can be caused by insects, severe weather, or harvest equipments<sup>30</sup>. It is common to find multiple *Pectobacterium* species together when symptoms are present<sup>17</sup>. The symptom of blackleg is black rotting of the stem from the mother tuber (Figure 4A). In addition, the leaves might be yellow and wilted. Only *P. atrosepticum* was believed to cause the bacterial blackleg however, *P. carotovorum* has also been shown to cause typical blackleg symptoms during potato plant infection<sup>31</sup>. The soft rot is usually associated with both *Pectobacterium spp*. When the bacterium is inside a tuber, it will decay the inside of the tuber, but not the tuber periderm. The symptoms of soft rot are tubers with a creamy maturing consistency that generally starts at the lenticels, stolons, or wounds (Figure 4B)<sup>4,17,30</sup>.

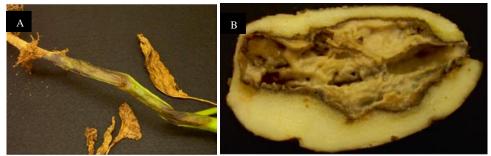


Figure 4: *R. Pectobacterium* spp. blackleg (A) and soft rot (B) symptoms<sup>28</sup>.

A third bacterial disease is the common scab. The pathogen is *Streptomyces scabiei*. It is present in valleys below 3,000 masl with an incidence that can rise to 90%<sup>4</sup>. *S. scabiei* enters through a natural opening such as the lenticels of tubers or wounds. If the pathogen enters, it causes lesions on the underground parts of a potato plant (roots, stolons, stems, and tubers) (Figure 5). It does not cause foliar symptoms, but the underground lesions can reduce the plant vigor<sup>4,17,27</sup>. Only young tubers are infected. Once the tubers are mature, the pathogens can no further enter them<sup>32</sup>.

A fourth bacterial disease is the *saq* ' $o^{4,33}$ . It is possibly caused by the association between an unknown phytoplasma and a virus (the potato leaf roll virus). The "*saq* 'o" is mainly present in valleys between 2,500 and 3,000 masl and reduces the yield between 20% to 40%<sup>4,33</sup>. The leaf symptoms are chlorosis or anthocyanescence, size reduction, and enrolment (Figure 6). Additionally, there is a high proliferation of the stolons, tubers, and roots. The flowers can be discoloured with a reduction of the peduncles and their size<sup>33</sup>. It is worth mentioning a final bacterial disease, locally called "*brotes grandes*" which is a 16SrI-B subgroup phytoplasma present in the province of Santa Cruz<sup>34</sup>.



Figure 5: S. scabiei tuber symptoms<sup>28</sup>.



Figure 6: "saq'o" leaf symptoms<sup>33</sup>.

To continue, a well-known disease is caused by the **oomycete** *Phytophthora infestans*. The disease is the late blight. The pathogen is characteristic of humid localities where precipitations are over 800 mm during the agricultural season<sup>29,35</sup>. It causes economic losses up to 100% when it is not controlled<sup>29,35</sup>. The primary inoculum sources are sporangia, zoospores, or mycelium subsisting on plant debris or tubers. They infect potato plants at any phase of the crop development starting the infection cycle<sup>35</sup>. Only mating type A2 was detected in Bolivia<sup>36,37</sup>. The symptoms are visible during the necrotic stage of *P. infestans* often where water accumulates at the leaf edges or tips, stems, and petioles (Figure 7A-B). Leaf lesions are irregular, water-soaked, and dark brown sometimes surrounded by a yellow halo and not limited. On the abaxial face surrounding the lesion, a white mycelium corresponding to the sporangiophores can form (Figure 7A). On the tubers, brown irregular marks can be observed going from the surface to the tuber medulla (Figure 7C)<sup>14,25,38</sup>.



Figure 7: P. infestans leaf (A), stem (B), and tuber (C) symptoms<sup>28</sup>.

In the dept. of Cochabamba, nine **fungi** are responsible for diseases reducing the potato production yield.

A first fungal disease is the early blight. The pathogen *Alternaria solani* causes moderate to high losses in the Carrasco Province (Figure 18, cf. page 25) between 3,000 and 3,500 masl and is present in the Colomi municipality<sup>4,29</sup>. *A. solani* conidia washed from the soil infect the leaf tissue through the intact epidermis, stomata, or wounds of older leaves from the flowering stage to the maturation stage<sup>17,29</sup>. On the leaves, the fungus forms brown necrotic spots from a few millimetres to two centimetres surrounded by a chlorotic border. The necroses are often delimited by the veins giving them an angular appearance (Figure 8A). In the spot, discontinuous and irregular rings can be observed (Figure 8B). The conidiophores present in the spot are straight and septate<sup>39</sup>. Tuber symptoms which are dark, slightly sunken rotting lesions can be present when the conidia are washed from the leaves (Figure 8C)<sup>17</sup>.



Figure 8: A. solani leaf (A and B), and tuber (C) symptoms<sup>28,40.</sup>

A second fungal disease is the Septoria leaf spot (*Septoria lycopersici* var. *malagutii*). It causes losses from 30% to 40% in cold and humid zones at 4,000 to 4,500 masl as in the Colomi municipality<sup>4,29</sup>. *S. lycopersici* var. *malagutii* generally starts to develop from pycnidiospores washed from plant debris on the soil surface or wild hosts<sup>41</sup>. The diseased leaves show dark brown spots often lighter at the centre, with an oval or round shape of 1 to 5 mm, with irregular rings (Figure 9A-B). The black pycnidia are scattered at the centre of the spot. No symptoms are observed on the aboveground organs<sup>42</sup>.



Figure 9: S. lycopersici var. malagutii leaf symptoms (A and B)<sup>40.</sup>

A third fungal disease is the Cercospora leaf blotch (*Cercospora solanicola* and *Cercospora solani*) which is an emerging disease identified in a few communities located between 3,000 and 3,400 masl<sup>4,29</sup>. *Cercospora* spp. affects both the leaves and the shoots. On the leaves are observed circular to irregular spots with just visible irregular, concentric rings (Figure 10A). The centre of the lesion can be covered by a white villosity formed by clusters of conidiophores and acicular conidia (Figure 10B)<sup>4,39,40</sup>.



Figure 10: Cercospora spp. leaf symptoms (A and B)<sup>40</sup>

A fourth fungal disease is the Phoma leaf spot of potato (*Stagonosporopsis andigena*) which is sometimes reported at altitudes from 2,000 to 3,500 masl<sup>4,43</sup>. No information is available on the spread of *S. andigena*<sup>43</sup>. The leaves, stems, and petioles of potato plants are affected. On leaves, the fungus causes small (from 2.5 mm to 1 cm in diameter) black and concentric lesions (Figure 11). The lesions are initially present on the lower leaves. With the development of the disease, the lesions reach upper leaves and may coalesce together. Severely affected leaves turn blackish. The stems and petioles develop elongate lesions. The pycnidia are light-coloured and embedded in the affected tissues<sup>42,44</sup>



Figure 11: S. andigena leaf symptom<sup>45</sup>

A fifth fungal disease is the grey mould (*Botrytis cinerea*). The grey mould is reported as an emerging disease between 1000-1500 masl in communities of the Colomi municipality. The grey mould does not cause significant losses<sup>40</sup>. *B. cinerea* generally infects the weak and senescent tissues. The foliar lesions are generally starting at the tip of the leaflets and develop along the central vein. The lesions present a chlorotic border plus they are brown to light grey or even darker if they are wet (Figure 12)<sup>46</sup>. Additionally, a characteristic grey mycelium can develop in favourable conditions, consisting of conidiophores and conidia of the fungus. It can also infect the inflorescence, fruits, and tubers<sup>40</sup>.



Figure 12: B. cinerea leaf symptom<sup>46.</sup>

A sixth fungal disease is the potato wart (*Synchytrium endobioticum*). The wart is found in almost all the potato productive areas<sup>4</sup>. Gandarillas mentioned to Torres (2002) that the tuber losses could reach 20% in Bolivia<sup>44</sup>. *S. endobioticum* infects all the aboveground parts except the roots with zoospores from the soil, human manipulation, or infected tubers<sup>4,17,47</sup>. Warts formed on belowground plant parts are compact protuberances. Tubers will develop white, purple, or green cauliflower-like warts that develop into galls (Figure 13). While on the aboveground, two main types of symptoms are observed: leaf malformations and shoot malformations<sup>4,17,47</sup>.



Figure 13: S. endobioticum tuber symptom<sup>28.</sup>

The silver scurf (*Helminthosporium solani*) was reported in the Palca community (Sacaba municipality) although symptoms are rarely present at harvest<sup>4</sup>. Most of the primary infection of *H. solani* happens before harvesting principally from conidia-infected potato seeds<sup>48</sup>. Jellies and Taylor (1977) mention infection of the progeny tubers as soon as tuber initiation begins<sup>49</sup>. During the storage, primary and secondary infections are possible<sup>48</sup>. The symptoms are irregular-shaped grey discoloration of the tuber periderm with the possible presence of black conidiospores at the border of the lesions (Figure 14)<sup>4,17,27</sup>. In addition, the lesions remain small in the soils but enlarge during storage<sup>48</sup>.



Figure 14: *H. solani* tuber symptom<sup>28</sup>.

An eighth fungal disease is the potato smut caused by *Thecaphora solani*. It is present between 2,500 and 3,000 masl<sup>4</sup>. The production losses do not exceed 10%<sup>4</sup>. *T. solani* infects the plant during sprout development from infected tubers or the soil<sup>4</sup>. The underground stems, the stolons, and the tubers develop hypertrophy and tumours with lobes (Figure 15). When they are cut, cavities in radial stripes containing the light brown spores are observed. The fungus does not cause symptoms in the vegetative system<sup>4</sup>.



Figure 15: T. solani tuber symptom<sup>4</sup>.

A final fungal disease is the black scurf (*Rhizoctonia solani* or *Thanatephorus cucumeris* during the sexual phase). The yield losses reported are between 30% and 40% in cold and humid climates between 4,000 and 4,500 masl<sup>4,29</sup>. *R. solani* infection can be tuber-borne or soil-borne. The infection starts early in the season generally causing an irregular emergence, wilting, and stunting in plants<sup>50</sup>. The apical leaves show curling and purple pigmentation (Figure 16C). The stems are depressed brown lesions with angular edges at the base (Figure 16B). During the sexual phase, a powdery, superficial, white mould appears on the stems just above ground level corresponding to the fungi basidiospores (Figure 16D). The formation of green aerial tubers in the basal and middle parts of the stem is possible. Regarding tubers, dark brown sclerotia develop on their surface later in the season (Figure 16A). Some tubers also present star-crossed skin crevices and trumpet-shaped holes as a symptom of *Rhizoctonia*<sup>17,27</sup>.

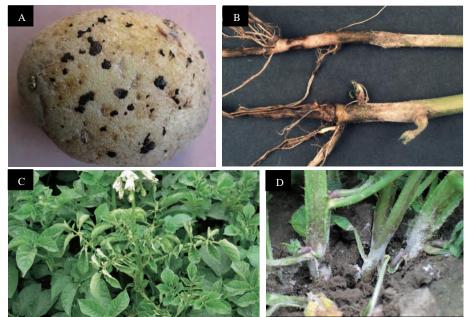


Figure 16: R. solani tuber (A), stem canker (B), leaf (C), and white stem (D) symptom<sup>28</sup>.

The powdery scab caused by *Spongospora subterranea* f. sp. *subterranea* is a **protozoon**. The powdery scab is found in almost all the potato productive areas<sup>4</sup>. *S. subterranea* zoospores infect roots, underground stems, and stolons of potato plants<sup>51</sup>. Fallon et al. (2016) mention that there is "an increasing body of evidence that the root infection stages of *S. subterranea* establish in young plants"<sup>51</sup>. On the roots and stolons, galls can form with sporosori inside it. On the tubers, it settles under the skin and develops light-coloured blisters that contain the sporosori (Figure 17). The lesions enlarge and darken before bursting and liberating the sporosori<sup>4,17,27</sup>.



Figure 17: S. subterranea tuber symptom<sup>28.</sup>

Lastly for the diseases, in addition to the common **viruses**, unique viruses only encountered in the Andean region are associated with potatoes. For Bolivia, these viruses are the potato leaf roll virus (PLRV), potato virus Y (PVY), potato virus X (PVX), potato virus S (PVS), Andean potato mottle virus (APMoV), Andean potato latent virus (APLV), the potato mop-top virus (PMTV), the tobacco rattle virus (TRV), the potato aucuba mosaic virus (PAMV)<sup>4</sup>. The potato virus A (PVA) is less studied in Bolivia<sup>4</sup>. The family, the genus, the vectors, and the main symptoms are presented in the Table 1 (cf. page 22).

The most damaging viruses worldwide are the PVY and the PLRV. Both viruses are responsible for tuber yield losses which can reach 80%. The PVX is also an important virus responsible for 10 to 40% of tuber yield losses in single infections. The yield losses can reach 80% in case of cross-infections with a potyvirus such as the PVY or the PVA. The PVS causes minor tuber yield losses in single infections. The PVS causes minor tuber yield losses in single infections. The PVA can also reduce the yield by 40% however its prevalence is less important than the PVY, PLRV, or PVX. The PMTV and the TRV are causing economic losses by affecting the quality of the tubers. The APLV and the APMoV are restricted to the Andean region<sup>17</sup>.

Garcia and Gandarillas (1992) identified that the most common viruses in the Cochabamba high Andean region (2900–3380 m) are PVX, APLV, APMoV, and PVY, and to a lesser extent PVS and PLRV<sup>52</sup>. Coca Morante (2021) confirmed that all these viruses were present in the Cochabamba department with the most common viruses being the PVX, PVY, and APLV. There are possibilities of crossed infection such as PVX + APLV and PVX + APMoV in the Cochabamba department or de PVY + PVX and PVY + PLRV for the La Paz department<sup>53,54</sup>.

In the Peruvian highlands (>3000 masl) the incidences reported are 30–82% for PVX, 20–50% for PVS, 4–15% for APMoV, and 2–6% for APLV. PLRV and PVY were usually only detected at 0–5% in potato plant material and, PMTV was uncommon<sup>55</sup>. While, for similar surveys carried out at lower altitudes in the Andean region (<3000 masl), PVY and PLRV were dominating<sup>17</sup>. No articles discuss which PVY strains are present in Bolivia. However, in Peru, the PVY<sup>O</sup>, PVY<sup>N</sup>, PVY<sup>C</sup>, and PVY<sup>NTN</sup> are confirmed<sup>36,56</sup>.

Virus	Genus, Family	Vector	Main symptoms	
Potato leaf roll virus	Polerovirus	Aphids	Primary infection presents lightening or	
(PLRV)			yellowing with a possible purpling and	
			light rolling of the upper leaves.	
			Secondary infection shows strong rolling	
		of the base leaves with possible a leathery		
			to papery leaf texture, short internodes,	
			possibly yellowing, and strong rolling of	
			the upper leaves. Erect growth habit is	
			present in both infection types.	
Potato virus Y	Potyvirus,		Different strains cause different	
(PVY)	Potyviridae		symptoms. The O strain causes mild to	
			strong mosaic on the leaves, systemic	
			necroses, wrinkle and rugosity of the	
		leaflets, dropping of leaves and nanism of		
		the plant in cases of severe infection. The		
		N strain causes mild mosaic and possibly		
		leaf necrosis. The NTN strain causes the		
		potato tuber necrotic ringspot disease.		
Potato virus A			The symptoms are mild mosaic of the	
(PVA)			leaves and with possibly wrinkling of the	
			leaves.	
Potato virus S	Carlavirus,	Contact,	There are no or few symptoms. Mild	
(PVS)	Betaflexiviridae	aphids	symptoms include light chlorosis of the	
			foliage, deepening of the veins, and	
			reduction of the leaves size.	
Potato virus X	Potexvirus,	Contact	There are no leaves symptoms or mild	
(PVX)	Alphaflexiviridae		mosaic and mottles, plus possible leaves	
			distortion.	

Table 1: Family, genus, vector, and the main symptoms associated with each virus<sup>17,28,33</sup>.

Potato aucuba			Yellow leaf flecking is accompanied with
mosaic			sometimes deformation and stunting.
virus (PAMV)			There are necrotic rings throughout the
			tuber flesh that are less clearly visible at
			the surface of the tubers than PMTV.
Andean potato	Comovirus,	Beetles	There are chlorotic blotches or mottles
mottle virus	Secoviridae		accompanied with leaves rugosity and
(APMoV)			plant dwarfism.
Andean potato	Tymovirus,		There are mild mosaic and chlorotic
latent virus (APLV)	Tymoviridae		netting of the minor veins on the leaves.
Potato mop-top	Pomovirus,	Spongospora	There are yellow colour blotching or
virus (PMTV)	Virgaviridae	subterranea	mottling, chlorotic V-shaped markings in
		f. sp.	leaflets, and extreme stunting of the
		subterranea	shoots. Tubers present necrotic rings, or
			lines throughout the flesh visible at the
			surface. Other possible symptoms on
			tubers are cracks and cross-linked skin.
Tobacco rattle virus	Tobravirus,	Nematodes	Symptoms include mottling and
(TRV)	Virgaviridae		deformation of the leaves, atrophy of the
			stems, characteristic pinch towards the
			end of the leaflet with edges of red-purple
			or yellow colour, possible tubers necroses
			sometimes visible on the skin surface.

### 2.4. Focus on Palca

Palca is a rural district of the municipality of Sacaba situated 40 km by road from the big city of Cochabamba and is in the central part of the Department of Cochabamba (Figure 18, cf. page 25). Palca is part of the Central Oriental Cordillera.

The proximity to the Andean-Amazonian transition zone generally determines the climate. For Palca, the north part where the altitude goes from 1,360 masl to 3,100 masl, the climate is defined as a humid subtropical climate where annual precipitations and average temperatures vary from 1,000 mm to 1,600 mm, and from 18 to 22°C respectively. Whereas the south part of the district, with an altitude between 3,400 masl and 4,400 masl is governed by a subhumid climate with annual precipitations and average temperatures varying from 600 to 1,000 mm, and 10 to 14°C respectively.

For both climates, the rain season goes from December to March and the dry season goes from April to September. The rain season comprises 75% of the annual precipitations<sup>14</sup>.

The mountains are strongly dissected with slopes between 30 and 60%. Three types of soils are encountered in the district. One of the three soil types, located at the west of the district, is lithic torriorthent. It is yellowish brown to pale brown, highly acidic, with an extremely low cation exchange capacity (CEC) and a moderate to medium texture. The second soil type, located in the north of the district is a humic dystustep soil type. This soil type is described with a loam texture, brown color, moderately acidic pH, and low CEC. Nitrogen, phosphorus, and potassium are found in high amounts. The third soil type is lithic cryorthent. This soil type is described with a moderately acidic pH, a very low CEC, and high amounts of nitrogen, phosphorus, and potassium<sup>14</sup>.

The predominant vegetations of Palca include several associations of graminoids grasslands and thickets. The shrubs are mainly dominated by tree species of the genus *Polylepis*. However, the action of humans transformed these forests into mainly grasslands and thickets. The forests are now reduced to inaccessible residual spots and act as refuges for endemic or endangered fauna and flora. At the northern end of the district, the predominant vegetation consists of sparse xeromorphic scrub and evergreen scrub<sup>14</sup>.

In Palca, the principal economic activity is agriculture with the principal cultures being potato, oca (*Oxalis tuberosa*), isaño (*Tropaeolum tuberosum*), papalisa (*Ullucus tuberosus*), oat (*Avena sativa*), barley (*Hordeum vulgare*), and green beans (*Vicia faba*). In the heights zone, potato cultivation is the most important with yields at 20 tons/ha, superior to the rest of the country. For example, in the *Kaluyo Chico* community, which is a community of Palca, in 2014, 27 hectares were assigned to potato production followed by 15 hectares of oats, 12 hectares of green beans, 8 hectares of Papalisa, and 8 hectares of Oca. Furthermore, the geographic localisation has allowed the cultivation of various varieties of native potatoes and Andean tubers. In fact, the cultural identity of Palca revolves around the potato. The livestock includes goats, llamas, cattle, equines animals, sheep, pigs, guinea pigs, and poultry all oriented mainly to self-consumption<sup>14</sup>.

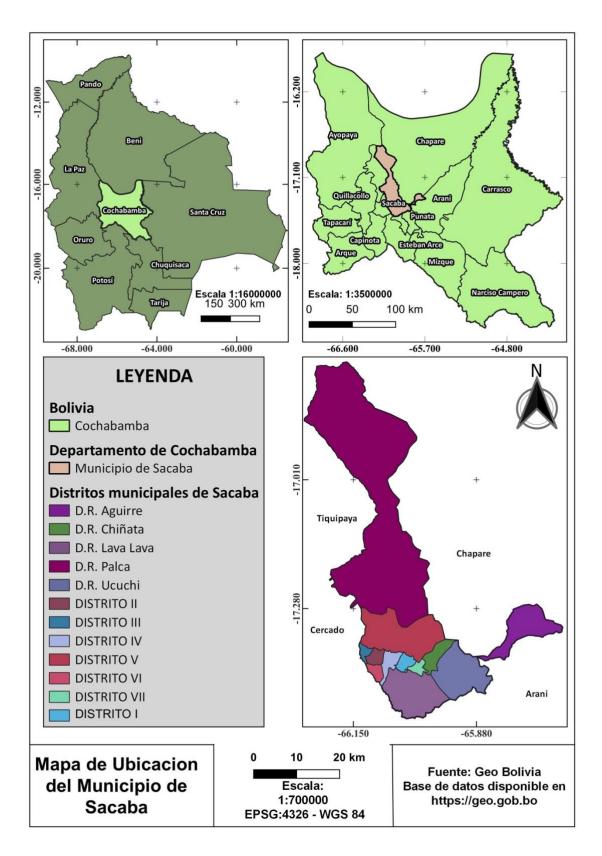


Figure 18: Political map of Bolivia, the municipality of Sacaba, and the district of Palca (Paco P, 2022).

# 3. Objectives

This master thesis is part of the project Root to Food realised by the Fontagro. As a brief reminder, the project goal is to provide good-quality seed tubers to the farmers of the municipality of Sacaba. The tubers are cleaned with in-vitro tissue culture by the Universidad Mayor de San Simón. By providing seeds with a good genetic and phytosanitary quality, the farmers of the municipality are also encouraged to adopt integrated disease and pest management strategies to reduce the use of phytosanitary products. The latter should positively impact the environmental and economic sustainability of the potato crops. In this context, to develop integrated diseases and pest management strategies, further information on the actual potato production system of the Sacaba municipality and the bio-aggressors associated with the potato crops in the municipality is important.

The study takes place in the Palca district which is part of the Sacaba municipality (Cochabamba department, Bolivia).

To do so, the first step requires understanding the potato production system at Palca. The farmers in the Andean region are dealing daily with agro-ecological conditions that can change in a matter of metres. This environment complexifies the potato production system. Realising an analysis of the agricultural practices carried out in the district of Palca is a necessary step to determine the good and bad practices realised by the farmers. It is useful to subsequently be able to propose appropriate solutions and alternative practices adapted to each case.

The second step aims to identify and quantify the bio-aggressors which are prejudicious to the potato production in the district. The bio-aggressors consider in this work are insect pests and bacterial, fungal, oomycete, protozoan, and viral diseases. The objective regarding the insect pests is to identify them and quantify their presence in the monitored plots. The objective regarding the disease is to identify the disease and quantify the level of infestation in the monitored plots. This information will provide an estimation of which bio-aggressors are more damaging in the district. It is complementary information that is helpful in the proposal of appropriate solutions and alternative practices for the farmers.

# 4. Materials and methods

# 4.1. Determination of the potato production system in Palca (Dept. of Cochabamba)

To determine the potato production system in the district of Palca of the Sacaba province (Department of Cochabamba) a survey was realised alongside farmers.

Three communities were selected to represent the overall potato production system. The communities of *Kaluyo chico*, *Písly*, and *Molinos* are located at different altitude levels from the highest to the lowest respectively. For each community, the survey was realised in groups of 5 to 10 persons.

The survey is presented in Annex 1 (cf. page 65). The questions were made according to the crop production system boundaries defined by Nemecek et al.  $(2014)^{57}$ .

### 4.2. Location of the plots and potato plant characteristics on each plot

The geographical coordinates, the altitude, the plot surface, the slope, potato varieties, and the potato plant growth stage (BBCH scale, Annex 2, cf. page 67) from the 21<sup>st</sup> of March to the 9<sup>th</sup> of April 2022 are presented in Table 2.

	Plot 1	Plot 2	Plot 3
Geographical	17° 13'10.32'' S	17° 14'40.73'' S	17° 15'6.93'' S
coordinates	66° 4'31.58'' W	66° 4'40.48'' W	66° 4'41.79'' W
Altitude	3,800 masl	3,900 masl	4,000 masl
Plot surface	3,425 m <sup>2</sup>	2,646 m <sup>2</sup>	1,496 m <sup>2</sup>
Slope	Moderately steep	Steep	Inclined
Potato varieties	Waych'a	Waych'a Pinta boca	Waych'a
Potato principal	Ripening of fruit and	Ripening of fruit and	Development to ripening
growth stage	seed to senescence (91)	seed to senescence (91)	of fruit and seed

Table 2: Geographical coordinates, altitude, plot surface, slope degree, potato varieties, and potato growth stage (BBCH scale) for the evaluated plots<sup>58</sup>.

A visualisation of the position and slopes of the plots in the district and the plots is displayed in Figure 19A and 19B.

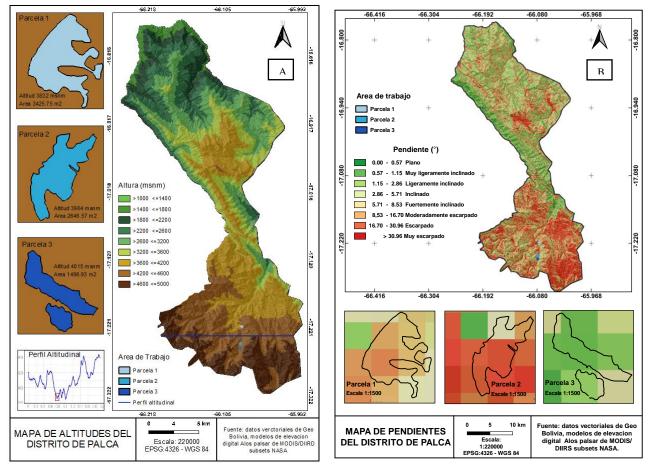


Figure 19: Cartographic position of the three plots per altitude (A) and slope (B) in the district of Palca (Paco P, 2022).

#### 4.3. Insect pest monitoring

The insect pest monitoring was realised by insect trapping and by visual observations of the plants and the tubers. The insect pests monitoring for the traps and the observations were realised weekly on the 27<sup>th</sup> of March, the 3<sup>rd</sup> of April, and the 9<sup>th</sup> of April of 2022.

#### 4.3.1. Insect trapping

The traps were positioned on the 21<sup>st</sup> of March 2022. Two types of traps were used to collect the insects: the von Moerick traps and pitfall traps (Figure 20). The traps were positioned in the centre of the plot to prevent any border effects. Per plot, three von Moerick traps were positioned on stakes at the heights of the foliage, in a triangle at 6 meters apart from each other. The von Moerick traps were filled with a mixture of water and inodorous soap to a line corresponding to a fourth of the container volume<sup>59–61</sup>. The pitfall traps aimed to capture weevils<sup>62</sup>. Three pitfall traps were positioned in-between plants at the top of the potato mounds, in a triangle at 6 meters apart from each other. The pitfall traps were filled with the same soapy mixture and covered with a wire mesh plus a roof<sup>62,63</sup>.

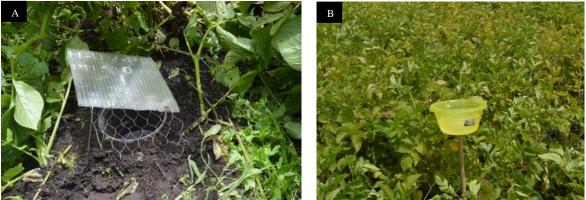


Figure 20: Pitfall (A), and von Moerick trap (B).

On collection days, the trap content was filtered onto a mesh and transferred into containers. The collection was conserved into a  $70^{\circ}$  denatured alcohol until further analysis.

#### 4.3.2. Visual observations

In addition to the insect trapping, visual observations of the plants were realised into 3 circles of a 1 m radius positioned randomly per plot<sup>59</sup>. The insects were captured and transferred into containers filled with 70° denatured alcohol until further analysis.

To assess the presence of larvae, three tubers per plant out of a total of 25 plants along a W-shaped path throughout the plot were also observed (cf. 2.4. Disease monitoring). The potatoes containing larvae were collected into bags for further analysis.

#### 4.3.3. Insect identification

The collected insects from trapping and visual observations were ordered. The pests were separated from the bulk samples of insects, counted, and classed taxonomically. They were identified up to the species level. The identification was realised visually with several identification keys up to the family taxonomic level under a binocular<sup>64–66</sup>.

#### 4.4. Disease monitoring

To avoid digging up the potato plants, the disease monitoring was realised on the shoot system and on the tubers. Each analysis was realised one week apart on the 27<sup>th</sup> of March, the 3<sup>rd</sup> of April, and the 9<sup>th</sup> of April 2022. Twenty-five plants were selected along a W-shaped path in each plot for the sampling<sup>67–69</sup>. The plant monitored were ten steps apart from one another. The entire shoot system and three potatoes were examined on each plant. The disease identification was based on symptomology caused by the bacteria, fungi, and viruses<sup>70</sup>.

Furthermore, for each disease, the collected data are the incidence and the severity. The incidence for each disease represents the percentage of diseased plants over the total number of plants observed. The severity is defined as the proportion of plant unit, usually relative to the total surface of plant tissue, that is affected by the disease<sup>71,72</sup>. Table 3 explains how the severity is evaluated for each disease apart from phytoplasma and viruses. Several standard area diagrams (SAD) were used to assess the disease severity. These SAD of the early blight, of the late blight, and of the powdery scab were used (Annex 3, cf. page 68)<sup>73–75</sup>.

Table 3: Severity evaluation realised per infectious agent.				
Infectious agent	Severity evaluation			
Ralstonia solanacerum	Percentage of wilted leaves on all plant <sup>76</sup> .			
Pectobacterium atrosepticum, Pectobacterium carotovorum var. carotovorum	Percentage of tissue of a tuber affected with soft rot <sup>77</sup> .			
Streptomyces scabies	Rating scale from 0 to 5 considering the scab lesions percent (% surface area), pitted lesions percent (% surface area) and depth of the pits (mm) on a tuber <sup>78,79</sup> .			
Alternaria solani	Percentage of the surface area with necrotic leaf tissue for all leaves of a plant <sup>80</sup> .			
Septoria lycoperici var. malagutii	Percentage of the surface area with necrotic leaf tissue for all leaves of a plant <sup>81</sup> .			
Cercospora spp.	Percentage of the surface area with necrotic leaf tissue for all leaves of a plant.			
Stagonosporopsis andigena	Percentage of the surface area with necrotic leaf tissue on all leaves of a plant.			
Phytophthora infestans	Percentage of the surface area with necrotic lesions on the leaves and shoots of the whole plant <sup>82</sup> .			
Botrytis cinerea	Percentage of the surface area with necrotic leaf tissue on all leaves of a plant.			
Rhizoctonia solani	Percentage of the surface area of a tuber covered with sclerotia <sup>83</sup> .			
Synchytrium endobioticum	Rating scale according to the number and sizes of warts on a tuber <sup>84</sup> .			
Spongospora subterranea f. sp. subterranea	Percentage of the surface area of a tuber covered with scab lesions <sup>73</sup> .			
Helminthosporium solani	Percentage of the surface area of a tuber covered with silver scurf lesions <sup>49</sup> .			
Thecaphora solani	Percentage of tuber tissues affected by the smut (no severity assessment encountered in the bibliography).			

Table 3: Severity evaluation realised per infectious agent.

Lastly, only symptom description was used to characterise the infection caused by the phytoplasma and the viruses.

## 4.1.Statistical analysis

Only disease severity values were statistically analysed using the software menu Minitab (version 21.1. 0). The data were treated following a linear generalised model (GLM). The factor considered is the plot (fixed and qualitative). The explicative variable is the severity.

The assumption for normal distribution was tested with the Ryan-Joiner test. The assumption for equal variance was tested with the Levene test. The assumption for normality was never verified therefore non-parametric Kruskal-Wallis tests were realised.

The null hypothesis was accepted when the p-value was superior to 0,05.

## 5. Results

#### 5.1. Results of the surveys

As mentioned previously, the production area of Palca is situated in the Central Oriental Cordillera between 1,360 masl and 4,560 masl. The mountains provide variability in the environmental conditions. Indeed, the variation in altitude influences factors such as temperatures, humidity, and precipitations. Moreover, different slope gradients are encountered in the district. These factors influence the crops and the livestock encountered as presented in Table 4.

Altitude		< 4000 masl	> 4000 masl	
Main crops	Potatoes and tubers	The farmers mentioned that it was most of the potato varieties for example Waych'a, Gollu, Double H, Canactillu, Pinta Boca, Zapallo, Olay, Oca, Isaño.	Luki (4 varieties), Pinco, Aquarniri	
	Other crops	Oats, barley, beans and for auto-consumption various vegetables	Oats, barley	
Main livestock		Cows, sheep, horses, pork, guinea pigs, llamas	Llamas	

Table 4: Crops and livestock depending on altitude.

The native potato varieties are cultivated for auto-consumption. The commercial potato varieties such as the Waych'a and the Holandesa are destined to farmer markets in Uspa Uspa, or Colcapirhua in the dept. of Cochabamba.

#### 5.1.1. The potato sowing periods

During the surveys realised, the farmers of the three communities mentioned three distinct sowing periods from June to November: June to July (*Mishka*) for the Písly community, August to September (*Chaupi Mishka*) for the three communities, and October to November (*Jatun Tarpuy*) for the three communities. The periods for soil preparation, planting, sowing, harvesting, and the time of the potato crop agricultural cycle associated with the sowing period are displayed in Table 5.

	Mishka	Chaupi mishka	Jatun tarpuy
Soil preparation	April, May	July, August	September, October
Sowing	June, July	August, September	October, November
Harvesting	January, February	March, April	May, June
Agricultural cycle	6 months	7 months	8 months

Table 5: Month of soil preparation, planting, sowing, harvesting, and the time for the culture cycle per sowing period.

#### 5.1.2. The agricultural practices of potato cultivation

Regarding agricultural practices, potato cultivation begins with the preparation of the soil. The use of mechanics depends on the access to the parcel and the degree of the slope. The soil preparation is realised a month prior sowing. The first step is to plough the field followed by the implementation of the furrows. Finally, the soil is fertilised with chicken manure. The quantity of chicken manure applied depends on the fertility of the soil (Table 6, cf. page 34).

The sowing is done when the soil is sufficiently humid generally two days after a rain. The sowing density is around 50 to 70 cm between hills and 30 to 40 cm between plants. A farmer of the *Písly* community said that he does 80 cm between the hills when plots are on a slope. Seed tubers have two origins. A part of the seed are certified tubers from the "*Unidad de Producción de Semilla de Papa*", and the other part of seed are small tubers from preceding productions.

There are two hillings realised during the cultivation. The first hilling is realised when most of the tubers sprouted (generally one month after the sowing). The right time to carry out the hilling depends on the rain and the humidity. The hilling is realised with a tool called the *Chuchuca*.

In addition to the fertiliser, the other inputs during the cultivation are fungicides and insecticides. Additionally, to the information provided in the Table 6 (cf. page 34), commonly, two or three applications of phytosanitary products are realised. The farmers of the *Molinos* communities realise two treatments of mixed fungicide and insecticide during the two hillings. The farmers of the *Kaluyo chico* community realise one insecticide treatment and two fungicide treatments. The first fungicide treatment is a preventive treatment against late blight. The second fungicide treatment is to "reinforce" the protection of the plant against late blight. A farmer of the *Písly* community said that he does 7, 5, and 3-4 applications of fungicides at low, intermediate, and high altitudes respectively. He does 4, and 3 applications of insecticides when insect pests are present. Herbicides are rarely used because at high altitudes the manual control of the weeds during the hillings is sufficient.

Lastly, the harvesting is realised manually.

	Fertiliser	Insecticide	Fungicide
Products used: company, active ingredients, additional information on the product	Chicken manure.	Karate zeon: Syngenta, 100 g/L lambda-cyhalothrine, large spectral foliar insecticide. Libertador: Nantong pest agrochemical limited company, lambda-cyhalothrine. Murille: no information found.	Ridomil: Syngenta, Metalaxyl 4%, Mancozeb 64%, systemic and contact fungicide for the control of late blight. Mancolaxil: UPL, Metalaxyl and Mancozeb. Maxim XL: Syngenta, 25 g/L Fludioxonil, 10 g/L Metalaxyl- M.
Objective mentioned by the producers	Fertilisation of the soil.	Control of the insect pests without further precisions ( <i>Molinos</i> and <i>Písly</i> communities). Control of the tuberworms ( <i>Kaluyo chico</i> community).	Control of the late blight ( <i>P. infestans</i> ) for the three communities.
Number of applications	1 application.	The number of applications mentioned varied between the communities.	
Time of the application	During the soil preparation.	During hillings and flowering of potat	o crop.
For fields that start to lose fertility, 5 bags of chicken manure are applied per charge of tuber seeds. For tired fields, 10 bags of chicken manure are applied per charge of tuber seeds. A bag corresponds to 1m <sup>3</sup> of manure. A charge corresponds to 112,5 kg of seeds.		The insecticide and the pesticide can be mixed and applied together. Information is lacking on the quantities and the application method.	

Table 6: Products used, objectives, number of applications, time of the application, practices associated with the fertiliser, insecticides, and fungicides.

#### 5.1.3. The crop rotation

The Figure 21 presents the crop rotation cycle of the Palca district. For the *Mishka* and *Chaupi Mishka* sowing, the crop rotation is only based on potatoes and a cereal which is oat (*Avena sativa*) or barley (*Hordeum vulgare*). Oat is more planted than barley. Potatoes are sowed in winter and cereals are sowed in summer. At high altitudes, the crop cycle is principally based on these two crops because of the cold weather. More rarely, beans (*Vicia faba*) can also be planted. At lower altitudes, in the rotation, oca (*Oxalis tuberosa*), isaño (*Tropaeolum tuberosum*), or Papalisa (*Ullucus tuberosus*) can replace potatoes. The plots are used if it gives proper potato yields without adding more than 10 bags of chicken manure per charge of seed. Over ten bags, the fields are left in fallow. A farmer of the *Písly* community said that he left his fields in fallow for 4 to 5 years.

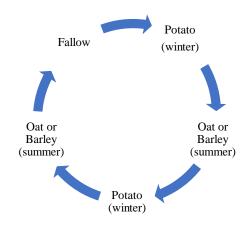


Figure 21: Crop rotation cycle in the Palca community.

#### 5.1.4. Pest risks associated with the potato crop

The principal insect pests are potato tuber moths and Andean weevils. Andean weevils are damaging at low altitudes while potato tuberworms are damaging at intermediate altitudes. Over 3800 masl, a very little number of insect pests is present. The diseases are the late blight, the "*manchas foliares*", the black scurf, and the potato wart. The late blight is a major problem. It was mentioned that the severity of the late blight decreases with altitude. The "*manchas foliares*" is a Spanish term that regroups the diseases provoked by *A. solani*, *S. lycopersici*, and *S. andigena* because of the similarity in the lesions on the leaves. The problems of "*manchas foliares*" are stronger at lower altitude.

The farmers of the *Molinos* community also mentioned a disease that they are calling the "*ismo*" which was described as holes generally in the eyes that mould during the stockage. They do not know what the agent of the disease is nor how to control it. They did not mention the quantity of losses occasioned by the "*ismo*".

#### 5.2. Insect pest results

A very little number of insect pests were present in the three fields examined. The insect pests encountered are *Phthorimaea operculella*, *Aphididae*, and *Curculionidae*. In addition, very little damage caused by the insect pests was observed on the leaves or the tubercules. Leaves damages (Figure 22) are only reported on two plants out of the 225 controlled. Only tubers of a single plant out of the 625 tubers controlled presented damages that could be associated to pest damages with certainty.



Figure 22: Leaf damage caused by insects (Bucher L., 2022).

#### 5.2.1. Phthorimaea operculella

The adult gelechiid moth is brownish grey with darker marbling and between 7 and 9 mm. The wings are in a roof-like shape. The forewings present fraying on the posterior edge but do not have a posterior cubital. The hindwings are trapezoidal with a bundle of bristles (frenulum). No tympanal organs on the abdomen base and the thorax. The larvae had a greenish white colour with a dark brown head and pinkish prothoracic shield. The pupa is reddish brown (Figure 23).



Figure 23: Adult (left), larvae (middle), and pupa (right) stages of P. operculella (Bucher L., 2022).

A total of 9 *P. operculella* are observed in two of the three plots (Table 7). In the plot 1, one moth is observed on the 27<sup>th</sup> of March 2022. In the plot 2, a total of 8 individuals are observed during the monitoring and all are present at the larvae stage in tubers of a single plant. Zero moths are observed in the third plot.

Undeterr	nined (und.)	corresponds	s to traps tha	t were vanda	alized (remo	oved) where	no counting	o counting was possible.			
		Plot 1		Plot 2			Plot 3				
	Yellow	Pitfall	Visual	Yellow	Pitfall	Visual	Yellow	Pitfall	Visual		
	water	trap	observ	water	trap	observ	water	trap	observ		
	trap		ations	trap		ations	trap		ations		
27/03/2022	0	0	1	0	0	0	0	0	0		
03/04/2022	0	0	0	0	0	0	0	0	0		
09/04/2022	0	0	0	und.	0	8	0	0	0		

Table 7: Number of *P. operculella* observed in the yellow water traps, the pitfall traps or by visual observations per plot. Undetermined (und.) corresponds to traps that were vandalized (removed) where no counting was possible.

#### 5.2.2. Aphididae

Two species of aphids were encountered: *Myzus persicae* and *Rhopalosiphoninus latysiphon* (Figure 25). The apterous *Myzus persicae* are light green to yellowish green, with quite long, clear slightly swollen cornicles and the frontal tubercles are converging. The apterous *Rhopalosiphoninus latysiphon* is olive green with black, shiny, swell cornicles and a plastron on the abdomen.

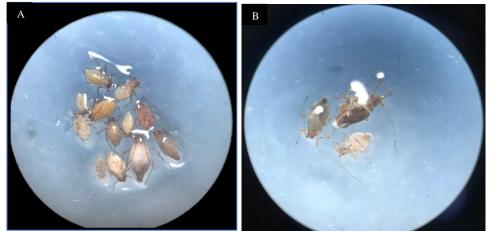


Figure 24: Observed Aphididae individuals, M.persicae (A), M.persicae and R. latysiphon (B) (Bucher L., 2022).

One individual out of the 16 was *R. latysiphon* whereas the others were all *M. persicae* (Table 8). No aphids were observed during the first collection on any of the three fields. Twelve aphids were trapped on the second plot during the third collection. Four aphids were trapped on the third plot during the second and third collection. No aphid was encountered by visual observation.

	Plot 1			Plot 2			Plot 3		
	Yellow	Pitfall	Visual	Yellow	Pitfall	Visual	Yellow	Pitfall	Visual
	water		observ	water		observ	water		observ
	trap	trap	ations	trap	trap	ations	trap	trap	ations
27/03/2022	0	0	0	0	0	0	0	0	0
03/04/2022	0	0	0	0	0	0	1	0	0
09/04/2022	0	0	0	und.	12	0	0	3	0

Table 8: Number of *Aphididae* observed in the yellow water traps, the pitfall traps or by visual observations per plot. Undetermined (und.) corresponds to traps that were vandalized (removed) where no counting was possible.

## 5.2.3. Curculionidae

The *Curculionidae* is a polyphaga beetle with a head extended into a rostrum, geniculated antennae with a long scape, and a differentiated club. The specimen measured 5 mm. The weevil species could not be identified (Figure 25). The *Curculionidae* was only observed once on the third plot of the last monitoring in the pitfall trap.



Figure 25: Curculionidae individual observed (Bucher L., 2022).

#### **5.3.** Disease results

### 5.3.1. Identified diseases

### 5.3.1.1. The Septoria leaf spot

For *Septoria lycopersici* var. *malagutii*, the foliage presented small brown round spots, with a "shiny" centre and a dark margin when oriented towards a source of light (Figure 26A). The lesions were not depressed in the leaf tissue. In the lesions, rings with pronounced concentric ridges are present and around the foliar lesions, a chlorotic hallow was sometimes observed (Figure 26C). Lesions were also present on the stems (Figure 26B).





Figure 26: Leaf lesions (A), stem lesions (B), and pronounced concentric ridges in a lesion (C) of the Septoria leaf spot (Bucher L., 2022).

#### **5.3.1.2.** The early blight

The early blight (Figure 27) caused by *Alternaria solani* presents very similar symptoms to the Septoria leaf spot. However, the lesions were brown without the "shiny" aspect of the centre, plus they were more delimited by the veins which gave them a more angular aspect (Figure 27A-B). The lesions were depressed in the leaf tissue. The chlorotic rings surrounding the lesions were generally stronger. In the lesions, the rings were irregular and not as pronounced (Figure 27D). In addition, spots were visible on the stems (Figure 27C). No tubers presented symptoms of the pathogen.

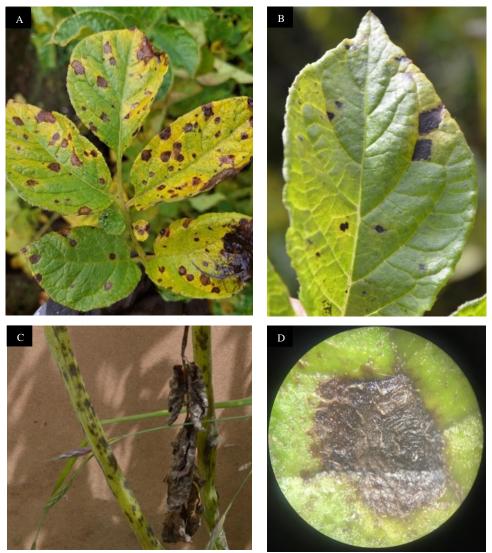


Figure 27: Leaf lesions (A), angular leaf lesions (B), stem lesions (C), and irregular rings in a lesion (D) of the early blight (Bucher L., 2022).

#### 5.3.1.3. The black scurf

Several symptoms caused by the *Rhizoctonia solani* were observed in the fields. To start, the apical leaves were curled upward with a pink to purple margin (Figure 28B). Numerous aerial tubers were generally present on the stems or at the ground level (Figure 28A). At the stem base, the white legs caused by the basidiospores were observed (Figure 28D). Finally, on the surface of the tubers was the black scurf caused by the fungi sclerotia (Figure 28C).



Figure 28: Aerial tubers (A), curled-up pinkish leaves (B), black sclerotia on tuber (C), and white mycelium at the stem base (D) of the black scurf (Bucher L., 2022).

#### 5.3.1.4. The late blight

For *P. infestans*, the leaves and the stems presented dark brown damp looking lesions where water tends to collect (Figure 29A-B). A white mycelium was observed at the periphery of lesions on the abaxial leaf faces (Figure 29C). No symptoms caused by the pathogen on the tubers were observed.



Figure 29: Stem lesions (A), leaf lesions (B and D), and white mycelium (C) of the late blight (Bucher L., 2022).

## 5.3.1.5. The powdery scab

Light-coloured blisters caused by *Spongospora subterranea* f. sp. *subterranea* were observed on the potatoes (Figure 30).



Figure 30: Light-coloured blisters of the powdery scab (Bucher L., 2022).

## 5.3.1.6. The potato wart

White and purple galls like cauliflower that could deform the tubers were observed in the fields (Figure 31). It is caused by *Synchytrium endobioticum*.



Figure 31: Purple and white galls of the potato wart (Bucher L., 2022).

### 5.3.1.7. The Cercospora leaf blotch

For *Cercospora* spp., the leaves presented a brown lesion with a white wooliness corresponding to the cluster of conidiophores (Figure 32). At the periphery of the lesion, irregular concentric rings are visible.



Figure 32: Leaf lesions of the Cercospora leaf blotch (Bucher L., 2022).

## 5.3.1.8. The PVY

For the PVY, the leaves presented vein necroses and necrotic spots on the reverse of the leaf (Figure 33A). Leaf deformations could accompany the necrotic spots (Figure 33B).

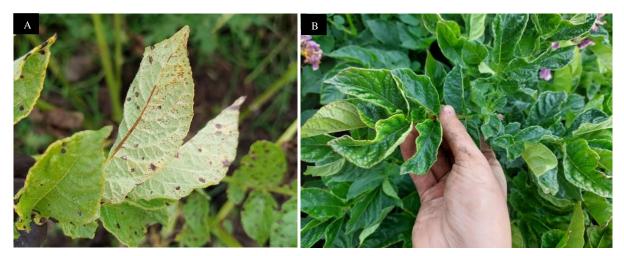


Figure 33: Vein necrosis on the leaves (A) and leaf deformations (B) of the PVY (Bucher L., 2022).

#### 5.3.2. Incidence and severity of the diseases

For each disease, the values of the observed incidence and severity for each plot during the three analyses are displayed in Table 9 (cf. page 47). To start, there are variations regarding the plant incidence, the tuber incidence, and the severity of each disease in-between plots. The first plot was the most affected by different pathogens while the second plot was the less affected.

The Septoria leaf spot was the most prevalent disease. It was present on all the plots with a plant incidence between 82% to 100%. The diseased plants were homogeneously distributed on the plot. The severity values were significantly different between the plots (p-value < 0.001). The severity on the first, second, and third plot are  $17\% \pm 12\%$ ,  $27\% \pm 18\%$ , and  $2.9\% \pm 3.3\%$  respectively. A significant difference was also observed between the plot 1 and the plot 2 (p-value < 0.001). The severity of the disease was the highest on the second plot. The severity was the smallest on the third plot. The Septoria leaf spot lesions are distributed homogeneously on the plant and for the two first plots. The lesions are only present on the bottom leaves for the third plot. On the leaves, the lesions are distributed homogeneously.

The early blight was present in two of the plots. On the first plot, the plant incidence was 13%. On the second plot, the plant incidence was 33%. The diseased plants were homogeneously distributed on the affected plot. The severity was low and significantly different (p-value < 0.05) between the two plots affected. The value corresponded to  $0.57\% \pm 1.8\%$  and  $0.76\% \pm 1.5\%$  for the first and the third plot respectively. The lesions were homogeneously distributed on the leaves.

The black scurf was registered in the three fields with a plant incidence going as high as 13% on the second plot. On the first plot, the plant incidence was only 4% every time which represents a single infected plant out of the 25 at each monitoring. On the third plot, the number of plants presenting symptoms were 4%, 8%, and 8% of the plant presenting symptoms on the first, second and third monitoring respectively. The tuber incidence was much lower and only tubers of the second plot presented symptoms (Table 9). The severity of the infected tubers was at a maximum of 10%. The sclerotia were homogeneously distributed on the tubers.

The late blight was present only on the first and the third plot. No plants presented symptoms on the second plot. Plus, no tubers presented symptoms of late blight for all plots. The oomycete was present on 4% of the plants and 5.3% of the plants on the first and the third plot respectively. It was only present on the third plot during the last monitoring. The severity was low and not significantly different (p-value > 0.05) on both plots presenting symptoms (Table 9). The severity of diseased plants was at a maximum of 15%.

The powdery scab was present in two of three plots. On the first plot, infected tubers were only seen during the last monitoring. Indeed, 23% of tubers and 11% of the plants were infected during the last monitoring. The infected tubers of the first plot were found from plants close to each other. On the second plot, 3.5% of the tubers were contaminated which represents one tuber contaminated out of the 75 observed during each of the first two monitoring and 8% of contaminated tubers on the last monitoring. The severity of diseased tubers stretched from a single lesion observed up to 30% of the surface tuber affected by lesions. No significant differences in the severity were observed between the two infected plots (p-value > 0.05).

The potato wart was only observed in the first field during each monitoring with a varying severity between "several medium-sized warts (> 10 mm)" and "large warts with a diameter of > 10 mm with disruption of tuber formation".

The Cercospora leaf blotch lesion was only seen once on a single leaf, on the first plot during the first monitoring.

Finally, for the PVY, the vein necroses caused were observed on 2.6% of the plants in the first field and 1.3% of the plants in the second field and the third field. It corresponds to two plants on the first field and one plant on the second and the third field presenting vein necrosis. Only the leaves on the third field were deformed.

Table 9: Plant incidence (% of plants presenting the disease in the sample, n=75); tuber incidence (% of tubers presenting the disease in the sample, n=225); severity of the Septoria leaf spot, early blight, late blight, and Cercospora leaf blotch (mean  $\pm$  stdev of the % of the leaf surface presenting lesions on the whole plant, n=75); severity of the black scurf, the powdery scab (mean  $\pm$  stdev of the % of the tuber surface presenting lesions on a tuber, n=225); severity of the potato wart (1= not affected, 5 = several medium-sized warts (> 10 mm), 6= several large warts, at least one of these being > 10 mm, and beginning deformation of the tuber, 7= large warts with a diameter of > 10 mm and disruption of tuber formation, n=225).

	Pl	ant inciden	ce	Tu	ber incider	nce	Severity		
	Plot 1	Plot 2	Plot 3	Plot 1	Plot 2	Plot 3	Plot 1	Plot 2	Plot 3
Septoria leaf spot	93%	100%	82%				$17\% \pm 12\%$	$27\%\pm18\%$	$2.9\%\pm3.3\%$
Early blight	13%	0%	33%	0%	0%	0%	$0.57\% \pm 1.8\%$	$0\% \pm 0\%$	$0.76\% \pm 1.5\%$
Black scurf	4%	13%	7%	0%	3.5%	0%	$0\% \pm 0\%$	$0.24\%\pm1.3\%$	$0\% \pm 0\%$
Late blight	4%	0%	5.3%	0%	0%	0%	$0.53\% \pm 2.4\%$	$0\% \pm 0\%$	$0.47\%\pm 2.0\%$
Powdery scab	3.5%	2.7%	0%	7.5%	3.5%	0%	$1.1\%\pm4.3\%$	$0.58\% \pm 3.6\%$	$0\% \pm 0\%$
Potato wart	11%	0%	0%	6.6%	0%	0%	1, 5, 6, 7	1	1
Cercospora leaf blotch	1.3%	0%	0%				$0.013\% \pm 1.1\%$	$0\% \pm 0\%$	$0\% \pm 0\%$
PVY (Vein necrosis)	2.6%	1.3%	1.3%						

## 6. Discussion

The potato is an essential element for food security in Bolivia. Families in the high Andes cultivate various varieties of potatoes with some varieties used for auto-consumption and other commercial varieties directed for the markets in the main cities of the country.

The production system of Palca is comparable to the rest of the Andean region in Bolivia. The variability of environmental conditions allows for different sowing periods of potatoes. The sowing period extends over nearly half the year from June to November. Thus, it offers a long period where fresh potatoes are available and gives access to better prices of potatoes on the market as mentioned by Terrezas and Garcias<sup>22</sup>. In the district, the three communities mentioned that they were sowing from August to September (*Chaupi Mishka*) and October to November (*Jatun Tarpuy*). Additionally, the communities at lower altitudes, such as the *Písly* community, start sowing earlier from June to July (*Mishka*). Indeed, the communities at lower elevations benefit from milder temperatures in winter and minor risks of frost compared to communities at higher elevations.

Regardless of the period of sowing, the yields of potato production are reduced because of bioaggressors associated with the crop.

The farmers mentioned several insect pests which are damaging to their production. The damaging pests are potato tuber worms and Andean weevils without furthermore precisions on the species. The pests observed are *Phthorimaea operculella* and aphids. No statistical analysis was realised because of their low presence. For P. operculella, there is no action threshold defined. However, the threshold of the guideline from the University of California is 15 to 20 moths per trap per night or a cumulative of 10 moths per trap per night<sup>85</sup>. No moths were caught in the traps. For the aphids, no action threshold for the damages provoked by probing the sap exists in Bolivia. In Belgium, the threshold is when 5 to 10 aphids are found on average per leaf (observed on the bottom, intermediate and upper leaf of 20 plants)<sup>86</sup>. Their quantity is below the threshold. Besides the insecticide treatment, the small number of insect pests observed can be caused by the altitude. Thus, a little number of pests was registered during this monitoring. This observation follows the comment of farmers that mentioned they were few pests over 3,800 masl. In Peru, the moth altitude dispersion is from 0 to 3,900 masl<sup>21</sup>. For the aphids, Kreuze et al. (2020) mentioned that there is a greater abundance of aphids under 3,000 masl in Ecuador<sup>17</sup>. Finally for the weevil, the pest species Rhigopsidius piercei, Premnotrypes latithorax, and Premnotrypes solariperda present protuberances on their abdomen which are not present on the captured specimen<sup>87</sup>. The specimen still felt worth mentioning because weevils are phytophages with numerous damaging species<sup>64</sup>.

The damaging diseases mentioned by the farmers were the late blight, the "*manchas foliares*" (that refers to *A. solani, S. lycopersici, and S. andigena*), the black scurf, and the potato wart. All these diseases were observed during the analysis. Additionally, on the monitored plots, the powdery scab, and the PVY were identified while no bacterial disease was identified in the monitored plot. It can be noted that the farmers only cited diseases presenting more distinctive and evident symptoms in comparison to viral diseases.

For the disease severity, it is important to acknowledge that the visual observations are subjective to the observer. The subjectivity might lead to inaccuracy of the severity, especially when realised by an inexperienced observer<sup>88</sup>. It is equally important to note that the evaluated data for the Septoria leaf spot and the early blight might present some inaccuracy due to the similarity of the symptoms. In-between the plots, different values regarding the incidence and the severity were registered. These differences can be the cause of various environmental variations that were not controlled. For the soil-borne pests, the different values of incidence in-between fields might be the result of differences in the concentration of the inoculum sources in the soils<sup>89</sup>. Another explanation for tuber-born pests is that more infected seed tubers were planted in the fields<sup>89</sup>. The development of the diseases is also influenced by various parameters such as the soils that were visually different (colour, depth, and texture were different) between the fields, the growth stage of the potato plants, the meteorological conditions like the precipitations, the humidity, the temperature, the wind that promote the pest development. Furthermore, all the diseases identified are common in the Andean region at an altitude varying between 3500 masl and 4500 masl with a constant high humidity caused by cloudiness and low temperatures<sup>29</sup>.

These environmental conditions are propitious to the development of *Septoria lycopersici* var. *malagutii*<sup>29</sup>. Indeed, the disease presented the highest plant incidence during the analysis. The severity varied between the plots. This difference in severity observed between the plots can be explained by a difference in the growth stage of the potato plants. Two of the plots with a close growth stage had a close severity  $(17\% \pm 12\%$  and  $27\% \pm 18\%$  for the first and second plots respectively). In comparison, the third plot with a younger growth stage presented lesions only on the lower leaves, and a lower severity (2.9%  $\pm$  3.3%) that was increasing over time. The incidence and severity reported in this work follow the trend observed by Coca Morante (2016) for leaves treated with Mancozeb and Clorotalonil<sup>72</sup>. A previous work mentions a severity of 60% in Ecuador<sup>90</sup>. The necroses of leaf tissue and defoliation caused by the infestation might reduce the assimilation area of the potato plant. Therefore, it may have a negative effect on tuber size and starch content. For this disease, during the production of 1999/2000 in the department of La Paz (4,290 masl) for the Waych'a paceña potato variety, a relation between severity (%) and yield (tons/ha) was developed. Following the trend line presented in the work, at the severity of 10%, the yield is about 15 tons/ha and at the severity of 30%, the yield is reduced to 11 tons/ha<sup>81</sup>.

The early blight presents close symptoms to the Septoria leaf spot. The Septoria leaf spot being dominant at this altitude (between 3,800 masl and 4,000 masl), the early blight values for the incidence and the severity are more probable to present some inaccuracy in comparison to the Septoria leaf spot. Coca Morante (2018) commented that the early blight does not seem to develop at altitudes over 4,000 masl certainly, due to the temperature being too low for its development (the optimal temperature corresponds to 20 °C)<sup>17,29</sup>. Thus, the observations appear to follow the idea of this comment. Indeed, the disease was only observed in two of the plots and the severity of the disease was low (0.57%  $\pm$  1.8% and 0.76%  $\pm$  1.5% for the first and third plots respectively). The disease is more prevalent between the altitudes of 2,900 masl and 3,500 masl<sup>29</sup>.

The black scurf was also mentioned as a problematic disease by the farmers. The incidence observed was higher on the second plot than in previous work done in Bolivia. Previous work realised in Bolivia assessed incidence values of 3 - 8.3% in 1985/86, 4.57 - 8% in 1986/87, and up to 8% in 1999/2000 in the La Paz department on the Cardinal, Alpha, and native varieties respectively<sup>50</sup>. To prevent the destruction of the potato plants, only the aerial parts of the plant and the tubers were observed. However, by doing so, some infected plants with a low severity that only present canker lesions on the underground stems, stolons, or roots can be missing in the counting. Additionally, since only tubers in the second plot presented sclerotia to assess the severity of the tubers, no statistical analysis was realised. For this, two explanations are plausible. Either the sclerotia were not formed yet or the observation of three tubers per plant was too little for this disease<sup>91</sup>. To continue with the severity, it can also be assessed with the canker lesions. Indeed, it better represents the damages provoked by the pest because the lesions may interfere with the normal movement of water and carbohydrates throughout the plant affecting the shape, size, and the number of tubers produced $^{92}$ . The aerial tubers, frequently observed during this study, are a sign of severe infection where the flow of carbohydrates might be reduced to the stolons. Consequently, these plants may present tubers with poor marketable quality<sup>92</sup>. For Bolivia, only yield losses were studied. Coca Morante (2019) reported yield reductions of 34 to 40% in 1999/2000 and 42% in 2016/2017 for diseased plants in comparison to healthy plants<sup>50</sup>.

Alongside the Septoria leaf spot was observed the late blight. This year, for the analysed fields, the late blight was not destructive to the production. Therefore, for this year, if yield losses are occasioned by the oomycete, they might principally be caused by the reduction of the green leaf area in plant tissues with necrotic lesions<sup>93</sup>. No infected tubers were observed. It does not conclude that there were no tubers lost in the fields this year because of the oomycete. Unfortunately, the farmers mentioned that the late blight is a major issue in their production with some years where *P. infestans* is devastating. In Bolivia, the damages caused to the leaves and the tubers can lead to the complete loss of the production when the disease is unproperly controlled because of a lack of information on proper management methods<sup>4</sup>. During the monitoring, an increase in the incidence was observed for the last analysis in the third plot.

Indeed, it is well known that by the polycyclic nature of the asexual cycle, the pest can spread rapidly, especially when the right environmental conditions for its development are met<sup>94</sup>. These conditions are mild temperatures with high humidity. The district of Palca corresponds to these conditions. Additionally, the Waych'a is a variety susceptible to the late blight but with a good capacity for regrowth.

The powdery scab is caused by the protozoon *Spongospora subterranea* f. sp. *subterranea*. The protozoon causes scab lesions on the tubers and root galls. During the monitoring, the tuber incidence was lower (7.5% and 3.5% for the first and the second plot respectively) than in previous works. Coca Morante (2014) determined a tuber incidence of 57% for the Waych'a variety in 2001/2002 and 2002/2003. Regarding the severity of the infected tubers, it is comparable to the previous results registered of 31% for the Waych'a variety in 2001/2002 and 2002/2003<sup>95</sup>. In this work, the infected tubers presented a severity from 1% to 30%. The plant incidence was low in both fields where it was present (3.5% and 2.6% for the first and second plots respectively). However, the pest was assessed only on the tubers and not the roots. The plant incidence could therefore be higher. For a long time, less emphasis was given to the root galls in comparison to the quality losses caused by the scab lesions on tubers. Different experiments reported reduced water uptakes and affected nutrient contents in shoots after *S. subterranea* inoculation<sup>51</sup>. These results emphasize the potential for *S. subterranea* to generally disrupt the growth and productivity of potato plants, and that the pathogen might affect potato productivity.

For the potato wart, the farmers mentioned its presence in their fields. The primitive fungus was only observed in the first field with a tuber incidence of 6,6%. The galls formed on the tubers were generally causing important tuber deformations. The fungus was reported to cause 20% tuber losses<sup>44</sup>. Even though the Waych'a is considered resistant to the fungus, the fungus is sometimes observed infecting this variety<sup>4</sup>. To which, PROINPA mentions that it may be due to another pathotype. The principal danger with the *S. endobioticum* is the persistence of resting sporangia that can survive more than 40 years without a host plant<sup>17</sup>.

Regarding the virus, the PVY was detected on the plots. Aphids which are the PVY vectors were observed on the plots during the monitoring. The vein necroses were observed on one or two plants per plot. In 2017, the PVX was more prevalent than the PVY in the Quechua region of Bolivia. During the same study, the incidence of viruses (PVX, PVY, PLRV, APLV, and APMoV together) was varying between 7% and 25% in the four communities monitored<sup>53</sup>. Consequently, it is probable that are greater incidence of viruses was present. Unfortunately, symptoms of mosaic were difficult to observe because the potato plants were at the senescent stage for the first and the second plots which led to the yellowing of the leaves (annex 4, cf. page 70). Furthermore, the identification only based on the symptoms is not

always possible for the viruses. For example, simple infection of the PVX only presents mild symptoms, or the only observation of mosaic symptoms does not allow for the identification of the virus species. Therefore, the symptomatologic identification should be accompanied by molecular analysis. To do so, a certain number of plants are systematically sampled. Once the virus genetic material is extracted, its detection can be realised with an enzyme-linked immunosorbent assay (ELISA). It can also be added that no symptoms of the potato mop-top virus were observed since powdery scab is a vector of the potato Mop-Top virus.

The latent infections were not tested in this work. Previous studies realised in the Cochabamba department detected the bacteria *Ralstonia solanacearum* in a latent stage over 2,800 masl<sup>4</sup>. Therefore, some plants might present pathogenic agents without expressing symptoms.

In Bolivia, the pests are mainly controlled with chemical phytosanitary products. However, the implementation of good agricultural practices and integrated pest management could reduce the development of pest populations. Consequently, it can help to reduce the use of pesticides which could help reduce the economic costs and minimize risks to human health and the environment<sup>96</sup>.

The first action line to consider is to prevent the development of pests and reduce the inoculum sources.

To do so, for the rotation cycles, the time is short between two potato cultivation. There is only a year between two potato cultivation. Such a short period may not be sufficient to properly reduce fungi inoculum sources in the soils. Indeed, a study realised in Canada demonstrated a reduction in the severity of soil-borne diseases with a rotation of three years (barley, red clover, and potato) in comparison with a rotation of two years (barley, potato)<sup>97</sup>. To continue, for *A. solani*, a study determined that at least two years in the rotation cycle are necessary to delay its onset and its development in the crop<sup>98</sup>. With problems of black scurf, at least three years are recommended<sup>85</sup>. For the powdery scab, at least three years are recommended by the University of California whereas Falloon (2008) recommends at least five years with a longer period likely to be necessary between two potato cultures<sup>85,99</sup>. For the late blight, at least one year is recommended between two potato cultures<sup>85</sup>. To properly manage the Septoria leaf spot, some information is lacking such as if the disease can also be seed-borne or how long the pest survives in plant debris<sup>41</sup>.

Before sowing, it is important to know the history of the plot regarding the pests<sup>100</sup>. Over 3,800 masl, the insect pests seem to be naturally controlled but at lower altitudes knowing the incidence of Andean weevils and potato tuber worms is important. For diseases, the same recommendation applies. Since the implementation of a larger rotation cycle for potatoes can be difficult in the short term. It is preferable to not sow potatoes in soils contaminated by the wart, the powdery scab, or the black scurf<sup>100</sup>.

For the black scurf, it is recommended to plant potatoes in fields where the disease has not been seen for at least three years<sup>101</sup>. For the wart, the recommendations are much larger because of the persistence of the resting sporangia. For example, the legislation of the European and Mediterranean Plant Protection Organization (EPPO) does not allow potato production for a period of 20 years with a safety zone where only the cultivation of resistant potato varieties is allowed in these areas (this legislation does not apply for Bolivia and is used as a reference)<sup>102,103</sup>. Indeed, for all diseases, sowing resistant varieties will reduce the damage provoked by the pests. The Waych'a variety is susceptible to the Septoria leaf spot, the late blight, and the powdery scab<sup>4,81,95,104</sup>. PROINPA has developed Waych'a varieties with horizontal resistance to the late blight<sup>4</sup>. On the other hand, the Waych'a presents resistance to the potato wart<sup>4</sup>. No information could be found on the resistance of the Pinta boca variety.

Obviously, the use of good quality tuber seeds that are free of diseases prevents their development. Coca Morante (2018) recommends the use of certified seeds in new soils used for crop production<sup>29</sup>. However, the only use of certified seeds is not economically accessible to small producers. After the harvest, the small tubers are collected to be sowed during the next production. To prevent the selection of bad-quality tubers, a positive selection can be implemented. To sum up, the farmers select the most vigorous and healthy potato plants without disease symptoms and during the harvest collect their tubers of the size of an egg which become tuber seeds for the next year<sup>101</sup>. Moreover, the use of good quality tuber would help in the reduction of viral secondary plant infections.

The distance between the rows and the plants follows the recommendations of the "*Unidad de producción de semilla de papa*"<sup>105</sup>. A good distance promotes better aeration between plants which reduces the accumulation of humidity and prevents the development of diseases such as the late blight and the Septoria leaf spot<sup>29</sup>. For *R. solani,* conditions for the rapid emergence of the potato plants reduce the risk of root and stem cankers<sup>106</sup>. To favour a rapid emergence, shallow planting and avoiding planting in cold, wet, and poorly drained soils are recommended. Well-drained soils are not only beneficial for preventing the development of *R. solani*, but also of *S. lycopersici, P. infestans, A. solani, S. subterranea*<sup>29,51,107</sup>.

For certain diseases, the use of organic amendments was shown to have a positive effect. Furthermore, plants that are not under stress conditions will be less susceptible to diseases. No manure from animals that consumed diseased potatoes should be incorporated into the fields. A study obtained a reduction of the powdery scab severity with organic amendments as a pre-planting treatment containing volatile fatty acids for soils with high levels of *S. subterranea* infestation<sup>99</sup>. For *R. solani*, it is possible to reduce the damages caused by the disease with amendments of mature compost<sup>108</sup>. The study was realised on cucumbers. Mature composts increase the activity of cellulolytic and oligotrophic actinomycetes. These latter are antagonistic soil microbes of *R. solani*<sup>108</sup>.

In fact, biological control strategies are recommended before the use of synthetic products in integrated pests management. In a thesis, parasitised larvae of *P. operculella* were observed in Bolivia<sup>109</sup>. No commercial biological control to propose which is already applied in Bolivia was found. Licensed products exist for the control of late blight (Planter-Box®, Polyversum®, and Polygandron®), early blight (Serenade<sup>®</sup> and Actisil<sup>®</sup>), black scurf (Proradix®)<sup>91,110,111</sup>. The potential of Plant Growth Promoting Bacteria could also help in the control. A *Bacillus amyloliquefaciens* isolated in Bolivia demonstrated a capacity to inhibit the infection of *R. solani*<sup>112</sup>.

When damages are still reaching the economic level, chemical control should be used. However, further knowledge is needed to be able to give feedback on how effective the control of the pests is in the district of Palca. In case the control of P. operculella becomes necessary, the insecticide treatment should be realised during the peak activity of the moth<sup>113</sup>. The peak activity is during the evening. Regarding primary infection of viruses, the strategy could be directed toward the vectors like aphids if primary infections are responsible for important economic losses. In that matter, synthetic insecticide uses are inefficient for aphid-borne viruses<sup>114</sup>. The use of hortical mineral oils has demonstrated better control of non-persistent aphid-borne viruses such as the PVY<sup>114</sup>. Unfortunately, aside from the risk of phytotoxicity, their expensive application costs make their use inadequate in the context of family farming<sup>114</sup>. For the control of fungal diseases, Cooke et al. (2011) mention that "it is important to know the efficacy and type of activity of the active ingredients to optimize the use of fungicides"<sup>115</sup>. "While the frequency and timing of fungicide applications depend on various factors such as the resistance of the varieties, fungicide characteristics, rate of growth of new foliage, weather conditions, and incidence of the disease in the region"<sup>115</sup>. No chemical control exists for the potato wart<sup>17</sup>. Therefore, good agricultural practices mentioned previously are important. To prevent any resistance development from the pests, phytosanitary products with the same action mode over a pest should not be used continuously. The producer should alternate between phytosanitary products with different action modes to control pests. Indeed, it was observed that the synthetic phytosanitary products mentioned, both for the insecticides and the pesticides, were generally composed of the same active molecules.

Finally, in Bolivia, no action thresholds exist for pest management. Defining action thresholds could help support the farmers in their decision processes. Indeed, the producers seemed disarmed when they were mentioning that some years, the late blight could completely ruin their production. To continue, there is also the disease *"ismo"* which was not identified because it was not observed during the monitoring. Regarding these aspects, the development of help supports for the problems encountered in their crop production or decision-making tools could help them to improve their yields and reduce the use of phytosanitary products.

# 7. Conclusion and perspectives

The study identified several action steps to look forward to the development of pests integrated management strategies in the district of Palca.

At the studied altitude, the insect pests did not cause significant damage to the production and their presence was below action thresholds preconised in integrated management. The insect pests identified are *Phthorimae operculella*, aphids (*Myzus persicae* and *Rhopalosiphoninus latysiphon*), and one *Curculionidae*. Over 3,800 masl, the systematic use of insecticide to control the insect pests could be questioned with rather the implementation of action thresholds. The study did not cover all the altitude levels of the district. Other studies should be realised at lower altitudes where the presence of insects is higher.

For the virus, the potato virus Y was present. The presence of other viruses like the PVX is probable and their detection with the support of molecular analysis should be the next step. The number of viral secondary infections should be reduced because of the project of the Fontagro to provide seeds of good sanitary quality. Regarding the fungal diseases, Cercospora spp. was not problematic. Septoria lycopersici var. malagutii is not highly studied however its presence is important in the studied plot. Further studies are needed to propose effective control strategies. For *Phytophthora infestans*, efforts are realised in Bolivia for its control due to the possibility of the total destruction of the production. Therefore, the efforts for the control of the late blight should be directed toward decision help supports and decision-making tools for the farmers of the district. For Rhizoctonia solani and Spongospora subterranea f. sp. subterranea, the use of fungicides is not directed toward their control. For these two diseases, the management strategy should be directed towards planting seeds free of the diseases, the reduction of the soil inoculum concentration in the fields with a longer period in-between two potatoes cultivation, and the use of resistant varieties. For plots affected by Synchytrium endobioticum, only the production of other crops or potato varieties resistant to the pathogen is recommended. Alternaria solani could be more damaging at lower altitudes. As for the insect pests, other studies realised at lower altitudes should be realised to determine the prevalence of the diseases in the district.

The work did not cover all the bio-aggressors. The cyst nematodes (*Globodera* spp.) were present in the fields. Their presence and their impact on the potato production of the district could also be an issue to investigate. Equally, the loss in the potato production occasioned by the bio-aggressors is not investigated in this work. It might present a possibility of future prospects to evaluate if the incidence and the severity reported by this work are responsible for important production losses. It could help in the implementation of action thresholds for the control of the bio-aggressors in the municipality.

# 8. Student personal contribution

The student wanted to develop competences in crop pests and their management which is not included in the teaching of the master Chemistry and bioindustries of bioengineering. Therefore, the student started with no experience and tried to fill its lack of knowledge by reading the courses on phytopathology, and insect pests during the first quadrimestre. The student developed the protocols in the fields to appropriately answer the problematic. The student learned Spanish to be able to interact with the potato producers of the region and to have access to the literature about the region. The student participated in the other ongoing projects in the centre of Nano-Biotechnology (UMSS, Cochabamba).

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# Annex 1. Questions of the survey

1. Characteristics of the physical space according to the altitude:

Ecological position		Valley	Summit
Altitude (masl)		<3000	>4000
Environmental conditions			
Main crops	Potatoes varieties		
	Other crops		
Main livestock			
Agricultural practices in re	lation with the soil use		

2. Surface cultivated by culture:

	Surface
Potato	
Oca	
Papalisa	
Isaño	
Beans	
Oats	
Barley, tarwi,	

3. The rotation cycles per production system:

	Mishka	Chaupi Mishka	Jatun Tarpuy
1 <sup>st</sup> year			
2 <sup>nd</sup> year			
3 <sup>rd</sup> year			
4 <sup>th</sup> year			
5 <sup>th</sup> year			
6 <sup>th</sup> year			

4. Agricultural practices with potato:

		Mishka	Chaupi Mishka	Jatun tarpuy
	Dates			
	Inputs: water, fertilizers?			
Soil preparation	Which one?			
I I I	Number of applications? When?			
	Energy carrier (mechanical, animals)			
	Machinery and tools			
	Dates			
	Conditions for planting			
Planting	Density (distance between plants + distance			
-	between rows)			
	Plot size			

Harvesting	date and practices		
Culture cycle (mo	onths)		
Culture management	Do you use fertilizers, herbicides, fungicides, insecticides, nematicides,? Which one? For what? Number of applications? When? Do you irrigate your fields? Is there any other work done on the potatoes field (e.g., manual weeding)? What are tools or machinery used?		-

#### 5. Risks and problems:

	Low altitude	Intermediate altitude	High altitude
Abiotic: frosts, hails, heavy rains, droughts?			
Insect pests			
Diseases			

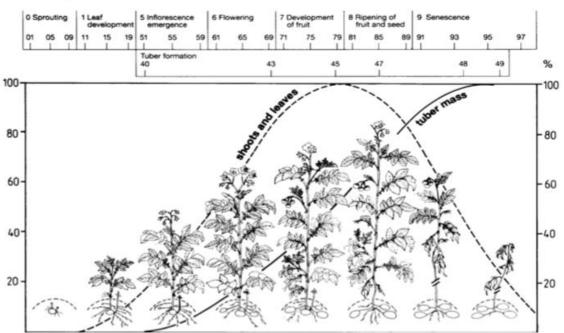
6. Potato production:

Is it sold? If yes, on which market? Is it for familial consumption? Is it for seed?

7. Potato seeds:

Where do you get the potato seeds? Are there yours from previous culture? Do you buy them? If yes, where? Do you exchange them with other farmers?

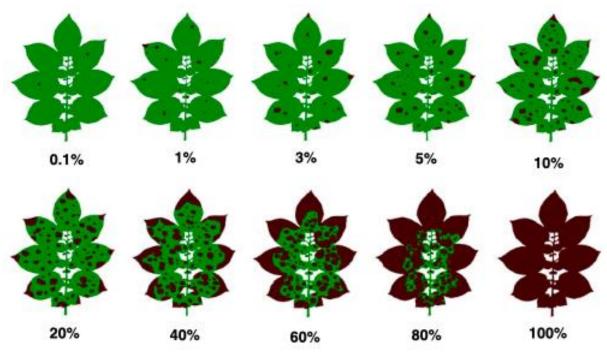
# Annex 2: Potato BBCH scale



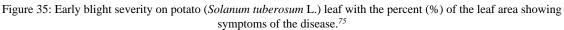
The 2-digit decimal code

Figure 34: Potato (Solanum tuberosum L.) 2-digit BBCH58.

Precision for the stage 91: Beginning of leaf yellowing.



Annex 3. Standard area diagrams



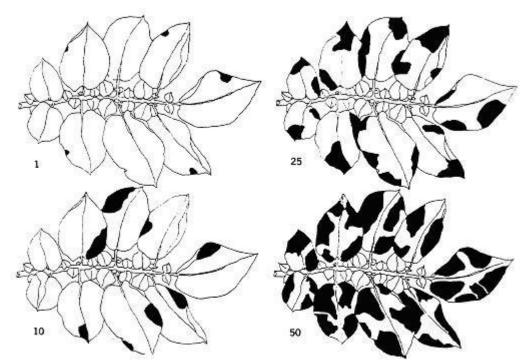


Figure 36: Late blight severity on potato (*Solanum tuberosum* L.) leaf with the percent (%) of the leaf area showing symptoms of the disease.<sup>74</sup>

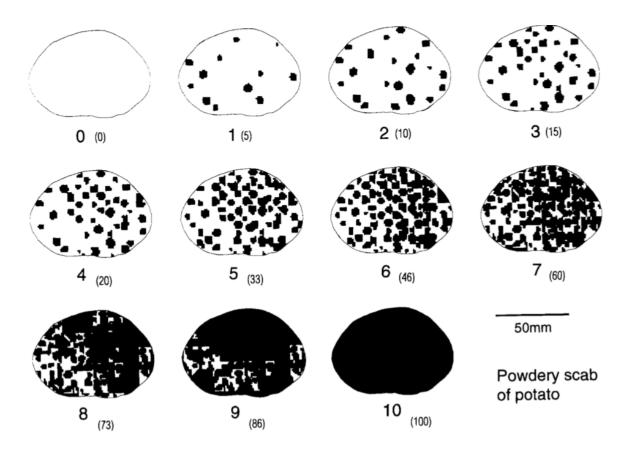


Figure 37: Powdery scab severity on potato (*Solanum tuberosum* L.) on tuber with the percent (%) of the tuber area showing symptoms of the disease.<sup>73</sup>

# Annex 4. Complementary pictures



Figure 38: Senescent stage of plants in plot 1 on 03/04/2022 (Bucher L., 2022).

	Severity (% of	Plot 1			Plot 2			Plot 3	
Plant	27-Mar	03-Apr	09-Apr	27-Mar	03-Apr	09-Apr	27-Mar	03-Apr	09-Apr
1	30	20	15	30	20	30	1	1	5
2	1	15	20	40	40	20	1	1	4
3	10	20	30	80	30	20	1	1	(
4	30	10	15	30	30	10	1	5	(
5	30	30	20	40	50	40	1	5	4
6	15	20	40	20	30	20	0	5	-
7	20	15	30	30	40	10	1	1	
8	30	20	10	40	20	10	0	5	1:
9	10	30	30	20	20	100	0	5	
10	40	20	10	40	20	30	1	5	1
11	10	10	1	20	30	40	0	5	
12	5	10	0	30	20	50	1	5	
13	0	5	5	10	30	40	1	0	
14	15	20	10	10	5	50	5	5	
15	20	30	20	20	10	20	5	1	
16	20	5	15	10	20	30	1	5	
17	0	30	5	40	5	20	1	5	
18	5	15	20	20	10	40	1	5	1
19	10	30	30	20	30	5	1	5	
20	0	1	10	30	100	30	1	5	
21	5	40	1	30	10	10	1	0	
22	20	30	0	40	10	20	1	0	
23	30	20	10	5	10	40	5	0	
24	40	1	5	10	20	5	10	5	
25	10	40	5	20	30	30	0	1	1

# Annex 5. Raw data of the severity

Table 11: Statistical analysis for the Septoria leaf spot.

Kruskal-Wallis test betw	veen p	lot 1,2, and 3		Kruskal-Wallis test on	the plot	t 1 and 2.	
Method	DF	H-Value	P-Value	Method	DF	H-Value	P-Value
Not adjusted for ties	2	113.02	0.000	Not adjusted for ties	1	13.45	0.000
Adjusted for ties	2	115.39	0.000	Adjusted for ties	1	13.91	0.000
Null hypothesis		All medians	1				
Alternative hypothesis	H <sub>1</sub> :	At least one	nedian is diff	erent			

		Plot 1		with testons) (	Plot 2	0		Plot 3	- I - · ·
Plant	27-Mar	03-Apr	09-Apr	27-Mar	03-Apr	09-Apr	27-Mar	03-Apr	09-Apr
1	0	0	5	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0
3	5	0	0	0	0	0	0	1	0
4	0	0	0	0	0	0	0	0	0
5	0	0	0	0	0	0	1	0	0
6	0	0	0	0	0	0	0	1	0
7	0	10	0	0	0	0	0	0	0
8	5	0	0	0	0	0	0	0	0
9	0	0	1	0	0	0	1	0	1
10	0	0	0	0	0	0	0	1	0
11	0	0	0	0	0	0	0	0	0
12	0	1	0	0	0	0	1	0	1
13	0	0	0	0	0	0	0	5	5
14	0	0	0	0	0	0	0	1	5
15	5	0	0	0	0	0	0	0	0
16	0	0	0	0	0	0	0	0	0
17	0	0	5	0	0	0	1	5	0
18	0	0	0	0	0	0	1	0	1
19	0	0	0	0	0	0	0	1	5
20	0	1	0	0	0	0	1	0	0
21	5	0	0	0	0	0	0	5	5
22	0	0	0	0	0	0	1	0	1
23	0	0	0	0	0	0	0	1	0
24	0	0	0	0	0	0	0	0	0
25	0	0	0	0	0	0	0	5	0

Table 12: Severity (% of foliar area covered with lesions) of the early blight for each plant (n=25) per date and plot.

Table 13: Statistical ar	alysis for the early blight.

Kruskal-Wallis test on the three plots.			Kruskal-Wallis test on the plots 1 and 3 (with sympto				
Method	DF	H-Value	P-Value	Method	DF	H-Value	P-Value
Not adjusted for ties	2	12.08	0.002	Not adjusted for ties	1	3.69	0.055
Adjusted for ties	2	30.44	0.000	Adjusted for ties	1	6.75	0.009
Null hypothesis Alternative hypothesis		All medians At least one	are equal median is diff	prent			

	Plot 1			(()))	Plot 2	0	Plot 3		
Plant	27-Mar	03-Apr	09-Apr	27-Mar	03-Apr	09-Apr	27-Mar	03-Apr	09-Apr
1	0	0	0	0	0	0	0	0	0
2	0	0	0	0	0	0	0	0	0
3	0	0	0	0	0	0	0	0	0
4	0	0	0	0	0	0	0	0	0
5	0	0	0	0	0	0	0	0	0
6	0	0	0	0	0	0	0	0	0
7	0	0	0	0	0	0	0	0	0
8	15	0	0	0	0	0	0	0	0
9	0	0	0	0	0	0	0	0	0
10	0	0	0	0	0	0	0	0	0
11	0	0	0	0	0	0	0	0	0
12	0	0	0	0	0	0	0	0	10
13	0	5	0	0	0	0	0	0	0
14	0	0	0	0	0	0	0	0	0
15	0	0	0	0	0	0	0	0	10
16	0	0	0	0	0	0	0	0	0
17	0	0	0	0	0	0	0	0	5
18	0	0	0	0	0	0	0	0	0
19	0	0	0	0	0	0	0	0	10
20	0	0	0	0	0	0	0	0	0
21	0	0	10	0	0	0	0	0	0
22	0	0	10	0	0	0	0	0	0
23	0	0	0	0	0	0	0	0	0
24	0	0	0	0	0	0	0	0	0
25	0	0	0	0	0	0	0	0	0

Table 14: Severity (% of foliar area covered with lesions) of the late blight for each plant (n=25) per date and plot.

Table 15: Statistical analysis for the late blight.

Kruskal-Wallis test on	the thre	e plots.		Kruskal-Wallis test on	the plo	t 1 and 3 (wi	th sympto
Method	DF	H-Value	P-Value	Method	DF	H-Value	P-Value
Not adjusted for ties	2	0.42	0.809	Not adjusted for ties	1	0.00	0.996
Tiot adjusted for des	-	0	12 0.007	Adjusted for ties	1	0.00	0.988
Adjusted for ties	2	4.13	0.127				
Null hypothesis	Ho: All medians are equal						
Alternative hypothesis	Hı: A	At least one n	nedian is different				

Plot	Date	Plant	Potato	Severity
1	09/04/2022	12	34	15
1	09/04/2022	12	35	15
1	09/04/2022	12	36	10
1	09/04/2022	13	37	20
1	09/04/2022	13	38	15
1	09/04/2022	13	39	15
1	09/04/2022	14	40	15
1	09/04/2022	14	41	15
1	09/04/2022	17	49	20
1	09/04/2022	17	50	30
1	09/04/2022	17	51	20
1	09/04/2022	18	52	20
1	09/04/2022	19	55	10
1	09/04/2022	22	64	1
1	09/04/2022	22	65	15
1	09/04/2022	22	66	10
1	09/04/2022	23	67	10
2	27/03/2022	8	22	10
2	03/04/2022	15	43	5
2	09/04/2022	1	1	30
2	09/04/2022	1	2	30
2	09/04/2022	1	3	30
2	09/04/2022	2	4	5
2	09/04/2022	3	9	10
2	09/04/2022	21	62	10

 Table 16: Severity (% of tuber area covered with lesions) of the powdery scab for each tuber per date and plot. Plants (n=25, per date per plot) and tubers (n=75, per date per plot) that are not present in the table did not present symptoms.

Table 17: Statistical analysis of the powdery scab.

Г

Table 18: Severity (% of foliar area covered with lesions) of the early blight for each plant per date and plot. Plants (n=25, per date per plot) that are not present in the table did not present symptoms.

Plot	Date	Plant	Severity
1	27/03/2022	12	0,1

Table 19: Severity (% of tuber area covered with lesions) of the black scurf for each tuber per date and plot. Present, the plant presented symptoms, but the tubers were not covered by sclerotia. Plants (n=25, per date per plot) and tubers (n=75, per date per plot) that are not present in the table did not present symptoms.

Plot	Date	Plant	Potato	Severity
1	27/03/2022	2	/	Present
1	03/04/2022	12	/	Present
1	09/04/2022	9	/	Present
1	09/04/2022	15	/	Present
2	27/03/2022	3	/	Present
2	27/03/2022	4	/	Present
2	27/03/2022	18	/	Present
2	03/04/2022	5	/	Present
2	03/04/2022	13	/	Present
2	03/04/2022	24	/	Present
2	09/04/2022	6	16	5
2	09/04/2022	14	40	10
2	09/04/2022	14	41	10
2	09/04/2022	20	58	5
2	09/04/2022	20	59	10
2	09/04/2022	24	70	5
2	09/04/2022	24	71	5
2	09/04/2022	24	72	5
3	27/03/2022	7	/	Present
3	03/04/2022	9	/	Present
3	03/04/2022	15	/	Present
3	09/04/2022	2	/	Present

Table 20: Severity of the potato wart (1= not affected, 5 = several medium-sized warts (> 10 mm), 6= several large warts, at least one of these being > 10 mm, and beginning deformation of the tuber, 7= large warts with a diameter of > 10 mm and disruption of tuber formation) per date per plot. Plants (n=25, per date per plot) and tubers (n=75, per date per plot) that are not present in the table did not present symptoms.

Plot	Date	Plant	Potato	Severity
1	27/03/2022	9	25	7
1	27/03/2022	13	37	5
1	27/03/2022	13	38	6
1	27/03/2022	13	39	5
1	27/03/2022	17	49	6
1	27/03/2022	17	50	6
1	27/03/2022	17	51	7
1	03/04/2022	19	55	6
1	03/04/2022	19	56	6
1	03/04/2022	19	57	5
1	03/04/2022	25	73	7
1	09/04/2022	1	1	6
1	09/04/2022	1	2	6
1	09/04/2022	6	16	5
1	09/04/2022	19	55	7

Table 21: Symptom description of plants infected by viruses per plot per day. Plants (n=25, per date per plot) that are not present in the table did not present symptoms.

Plot	Date	Plant	Symptom description
1	27/03/2022	/	One leaflet with vein necrosis
1	03/04/2022	9	Several leaflets with vein necrosis
2	03/04/2022	16	Several leaflets with vein necrosis
3	27/03/2022	/	One leaflet with vein necrosis, leaf deformations