

2023 Project Summary: Using Genetic Tests to Confirm Herbicide Resistant Weeds in Ontario Crops

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Since 2016, this project has developed 24 genetic quick tests (more in progress) to assist in identifying herbicide resistance in 14 weed species and confirmed 207 new cases of herbicide resistance in Ontario crops. These tests deliver a diagnostic and a recommendation to the grower within the same growing season. Traditional resistance testing in the greenhouse can take from three months to a year to get results back to growers. Now, leaf tissue instead of seed is collected. DNA is extracted from the leaf tissue to determine if there is a change in the sequencing resulting in a mutation making the plant resistant.

Tests have been developed to differentiate between Brassica and Amaranthus (pigweed) species. Tests differentiating pigweed species have been instrumental in confirming new cases of waterhemp in Ontario, Manitoba and Quebec. Once confirmed, the waterhemp is tested for Groups 2, 5, 9 and 14 resistances. Waterhemp has been found in 18 Ontario counties.

Table 1. Genetic Tests Currently Utilized by Harvest Genomics

| Weed Species | Herbicide Group | Resistance & Tests |
|--------------------------|-----------------|---|
| Large crabgrass | 1 | Metabolic: ACCase gene amplification |
| Common chickweed | 2 | Target-site (P197Q & unpublished) |
| Common ragweed | 2 | Target-site (W574L) |
| Eastern black nightshade | 2 | Target-site (A205V) |
| Giant foxtail | 2 | Target-site (unpublished) |
| Giant ragweed | 2 | Target-site (W574L) |
| Pigweed spp. | 2 | Target-site (S653N & W574L) |
| Common ragweed | 5 | Target-site (V219I) |
| Giant ragweed | 5 | Target-site (V219I) |
| Lamb's-quarters | 5 | Target-site (S264G) |
| Pigweed spp. | 5 | Target-site (A251V, S264G, V219I & F274L) |
| Brassica spp. | 9 | Presence of transgene |
| Canada fleabane | 9 | Target-site (P106S) |
| Common ragweed | 9 | Thr102Ile, Ala103Val, Pro106Ser sequencing assay |
| Italian ryegrass | 9 | Pro (CCA) to Ser (TCA) mutation at Codon 106 in EPSPS |
| Waterhemp | 9 | Metabolic: EPSPS gene amplification |
| Common ragweed | 14 | MAPAQ mutation R98L |
| Pigweed spp. | 14 | Target-site (Δ G210 in PPX2L) |
| Amaranthus spp. | - | Species identification |
| Brassica spp. | - | Species identification |

Note: New test from MAPAQ highlighted in yellow

Note: Amaranthus spp. Includes green pigweed, redroot pigweed and waterhemp

In 2018, the protocols for these tests were shared with the Pest Diagnostic Lab of the Quebec Ministry of Agriculture, Fisheries and Food (MAPAQ) and the weeds lab of AAFC's Harrow

Research and Development Centre as a pilot project and made available to extension personal in Ontario and Quebec to submit samples, providing the diagnostic service to growers.

In 2019, all samples were sent from Ontario to the Pest Diagnostic Lab of the Quebec Ministry of Agriculture, Fisheries and Food (MAPAQ), whom provided the testing for free. In 2020, MAPAQ could no longer accept samples from out of province.

In 2020, Harvest Genomics www.harvestgenomics.ca signed an agreement with AAFC to obtain the protocols and started to provide the service to Ontario growers for a fee. In 2023, the partners in Harvest Genomics disbanded. TurnKey Genomics was then formed www.turnkeygenomics.ca and they obtained a licensing agreement from AAFC to provide the genetic testing service in Ontario for this project.

Results

Table 2. 2023 Results in Ontario to October 27

| Crop | Weed | Herbicide Group | Total Fields | Positive Tests | % |
|---------------|------------------|-----------------|--------------|----------------|-----------|
| Carrots | Pigweed spp. | G5 | 15 | 9 | 60 |
| | | G5/14 | | 6 | 40 |
| Corn | Common ragweed | G2/G5 | 2 | 2 | 100 |
| Corn | Green foxtail | G2 | 1 | 0 | 0 |
| Corn | Pigweed spp. | G2/G5/G14 | 1 | 0 | 0 |
| Corn | Waterhemp | G2/G9 | 2 | 1 | 50 |
| | | G2/G9/G14 | | 1 | 50 |
| Grapes | Italian ryegrass | G9 | 1 | 0 | 0 |
| IP Beans | Common ragweed | G2/G5 | 4 | 2 | 50 |
| | | G2/G5/G14 | | 2 | 50 |
| IP Beans | Pigweed spp. | G2 | 1 | 1 | 100 |
| Popcorn | Lamb's-quarters | G5 | 1 | 0 | 0 |
| Potatoes | Lamb's-quarters | G5 | 1 | 0 | 0 |
| Potatoes | Pigweed spp. | G5 | 1 | 1 | 100 |
| Soybeans | Canada fleabane | G9 | 1 | 1 | 100 |
| Soybeans | Common ragweed | G2/G5 | 11 | 5 | 100 |
| | | G2/G5/G14 | | 6 | 100 |
| Soybeans | Pigweed spp. | G2 | 2 | 1 | 100 |
| | | G2/G5/G14 | | 0 | 0 |
| Soybeans | Waterhemp | G2/G14 | 12 | 3 | 25 |
| | | G2/G9/G14 | | 5 | 42 |
| | | G9/14 | | 3 | 25 |
| | | G5/G9/14 | | 1* | 8 |
| Tomato | Pigweed spp. | G5 | 1 | 1 | 100 |
| Unknown | Waterhemp | G2/G14 | 1 | 1 | 100 |
| Wheat | Waterhemp | G2/14 | 2 | 1 | 50 |
| | | G2/G9/G14 | | 1 | 50 |
| White beans | Common ragweed | G2/G5/G14 | 1 | 1 | 100 |
| White beans | Lamb's-quarters | G5 | 1 | 0 | 0 |
| Totals | | | 61 | 55 | 90 |

Note: Pigweed spp. includes green pigweed and redroot pigweed.

Note: The above data does not include 55 samples for waterhemp confirmations and resistance testing completed for Manitoba.

*Only one field has been found with G5 resistance at this time. There are 2 mechanisms of G5 resistance - target-site and metabolic. Seed will need to be collected and dose response experiments will need to be conducted on all fields to determine if G5 metabolic resistance and G27 resistance is present. There is no genetic test developed for G27 resistance yet. One is being worked on by Quebec researchers.

Significant Results:

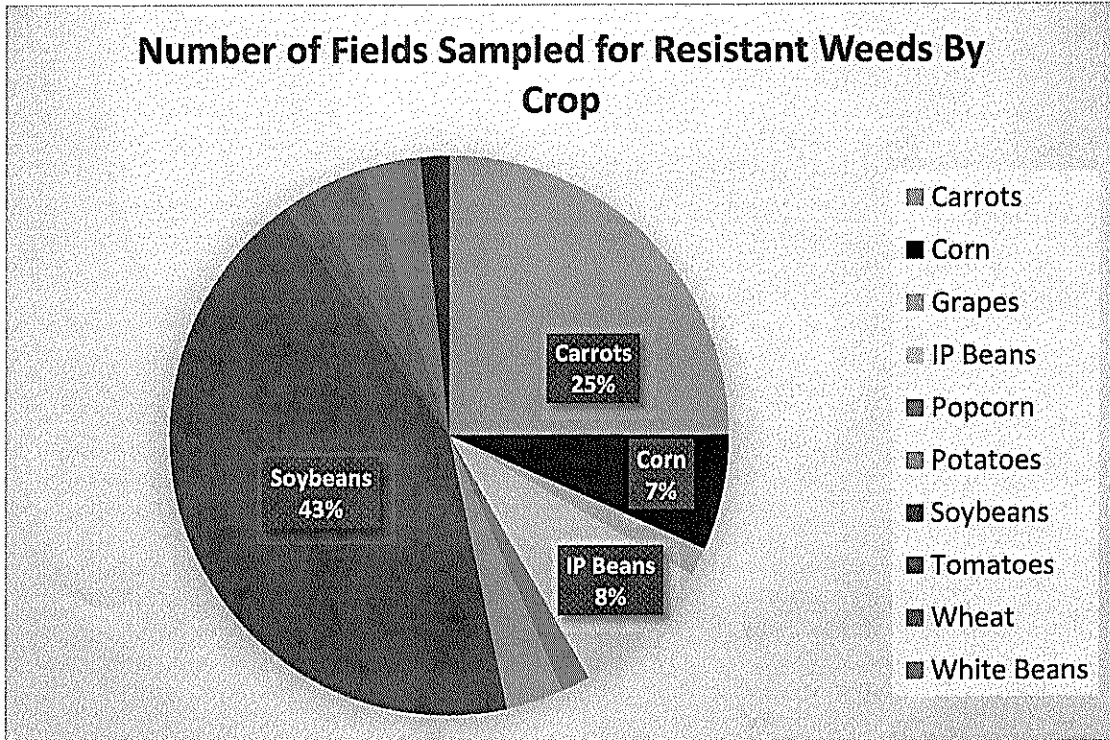
- Five-way resistant waterhemp to herbicide groups 2, 5, 9, 14 and 27 has been confirmed in 7 counties in Ontario - Chatham-Kent, Essex, Elgin, Lambton, Middlesex, Northumberland and Stormont, Dundas & Glengarry.
- Waterhemp has been confirmed in 18 counties in Ontario (Brant, Bruce, Chatham-Kent, Dufferin, Elgin, Essex, Haldimand, Huron, Lambton, Leeds and Grenville United Counties, Middlesex, Niagara, Norfolk, Northumberland, Ottawa, Stormont, Dundas and Glengarry, Wellington and Wentworth). No new counties were found this season.
- Over the course of this study, multiple resistant waterhemp has been confirmed in asparagus, corn, peppers, soybeans, sweet corn, wheat and white beans in Ontario. Wheat is new in 2023.
- In 2023, 100% of waterhemp confirmations were G14 resistant compared to 67% G9 resistant.
- Multiple resistant pigweed species (green pigweed and redroot pigweed) are commonly found in many horticulture crops for example: G2/G5 in pumpkins, potatoes, strawberries, sunflowers and tomatoes and G5/G14 in carrots.
- 33% of all pigweed spp. samples were multiple resistant to G5/G14 herbicides. All samples came from carrot fields.
- All common ragweed samples were multiple resistant. With 50% resistant to G2/G5 and 50% resistant to G2/G5/G14 herbicides. The common ragweed samples came from corn (2), IP beans (4), soybeans (11) and white beans (1).
- Three-way resistant common ragweed to herbicide groups 2, 5 and 14 has been confirmed in Bruce, Lambton and Prescott and Russell counties.
- Continued documentation of Canada fleabane resistant to G9 in apples, blueberries, carrots, grapes, onions, pumpkins and strawberries.
- Amaranthus species identification showed that waterhemp is often confused with green pigweed and tumble pigweed.

This testing has been instrumental in documenting new cases of herbicide resistant weeds. In 2023, 90% of the fields tested in Ontario were resistant to at least one herbicide group. Eighty percent of these fields were 2-way or 3-way resistant. Once confirmed producers were provided the resistance profile enabling a change in management to mitigate spread. Producers, agri-business and consultants that participated in the project were pleased with the timely results, welcomed the in-season management recommendations and highly value this service. For the most up to date herbicide resistant weeds information, visit our herbicide resistant weeds database on the Ontario Crop Protection Hub: [Herbicide Resistant Weeds - Database and Maps \(gov.on.ca\)](https://www.gov.on.ca/ocph/herbicide-resistant-weeds-database-and-maps)

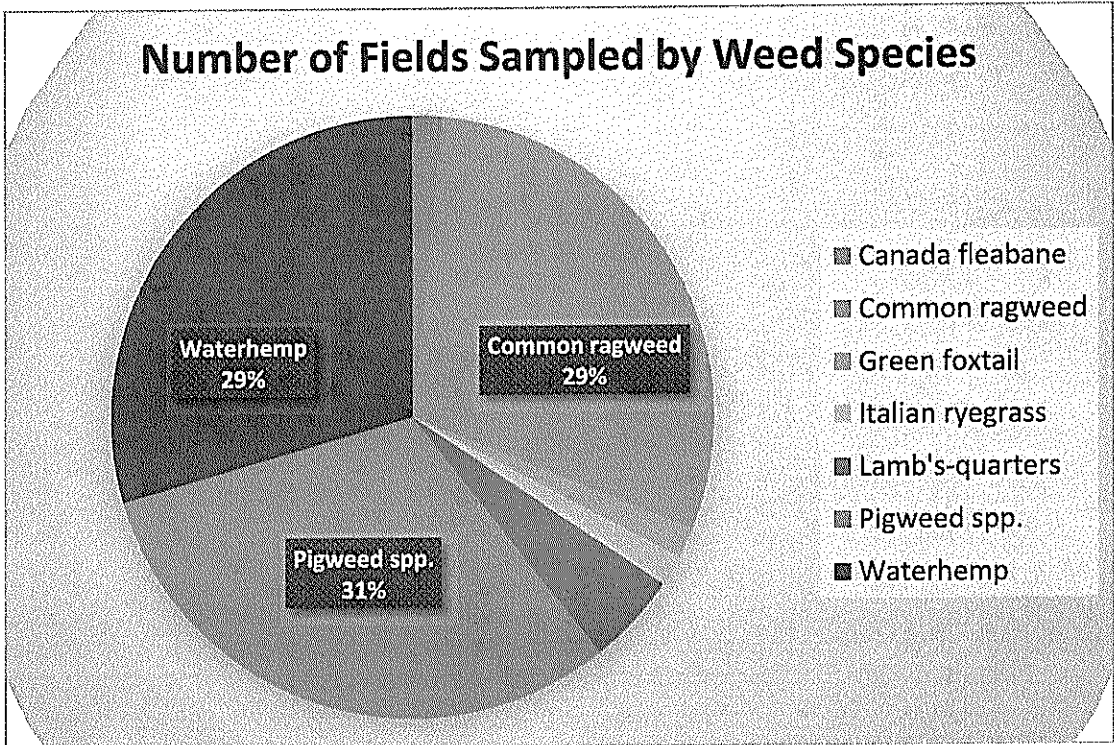
There are many more undocumented cases of herbicide resistant weeds in Canada. The resistance mechanism is unknown for most of them. The major concern is their distribution and economic impact for producers. Knowing where resistant biotypes are located will improve management and maintain the longevity of our crop protection tools.

Project partners include: AAFC, AAFC-PMC, Bayer Crop Science Inc., BASF Canada, FMC Canada, FVGO, MAPAQ, OAG, OFVGA, OPVG and Syngenta Canada Inc.

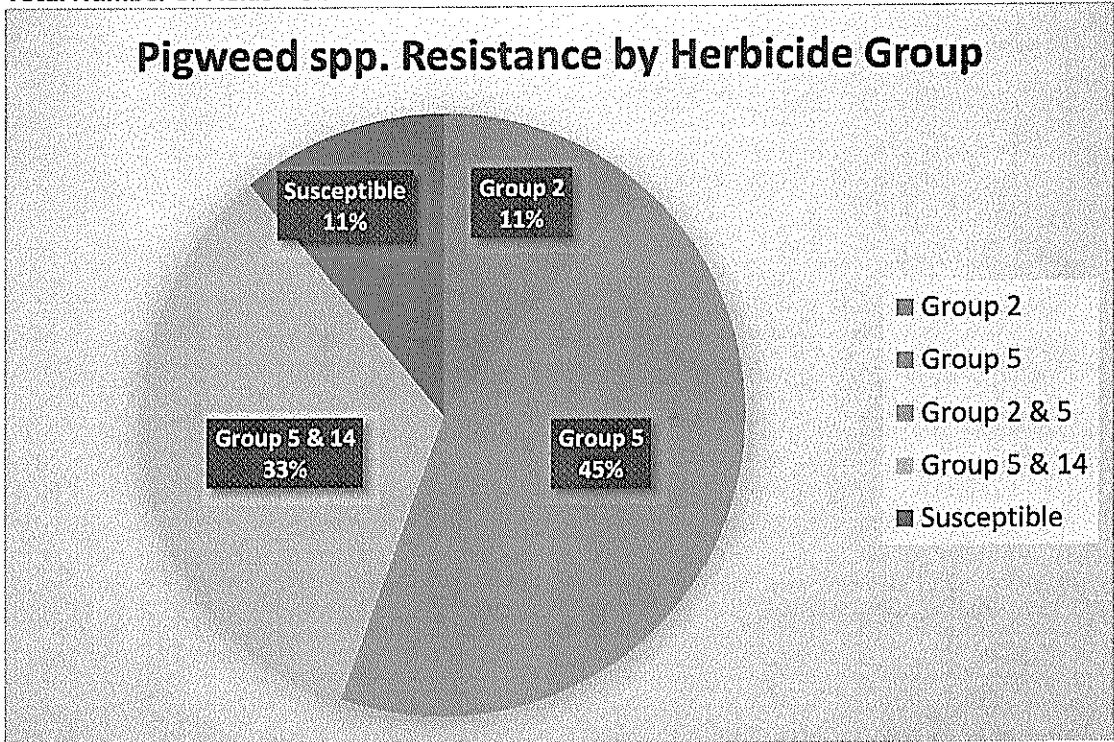
APPENDICES



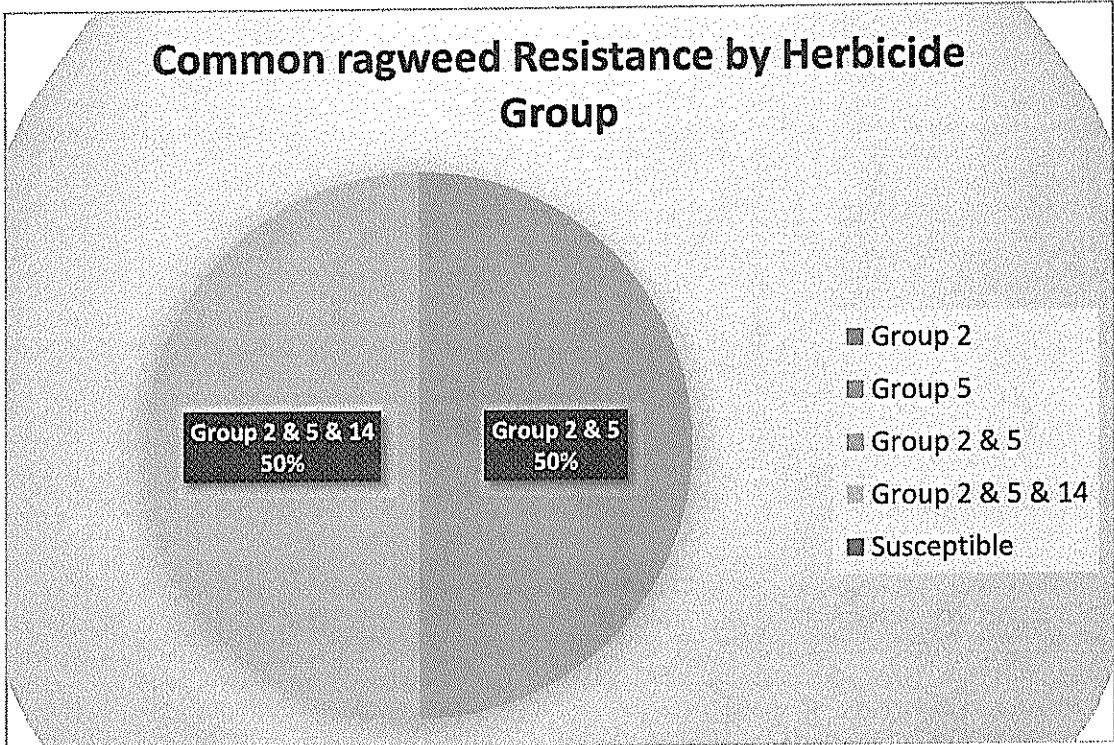
Total Number of Fields = 61



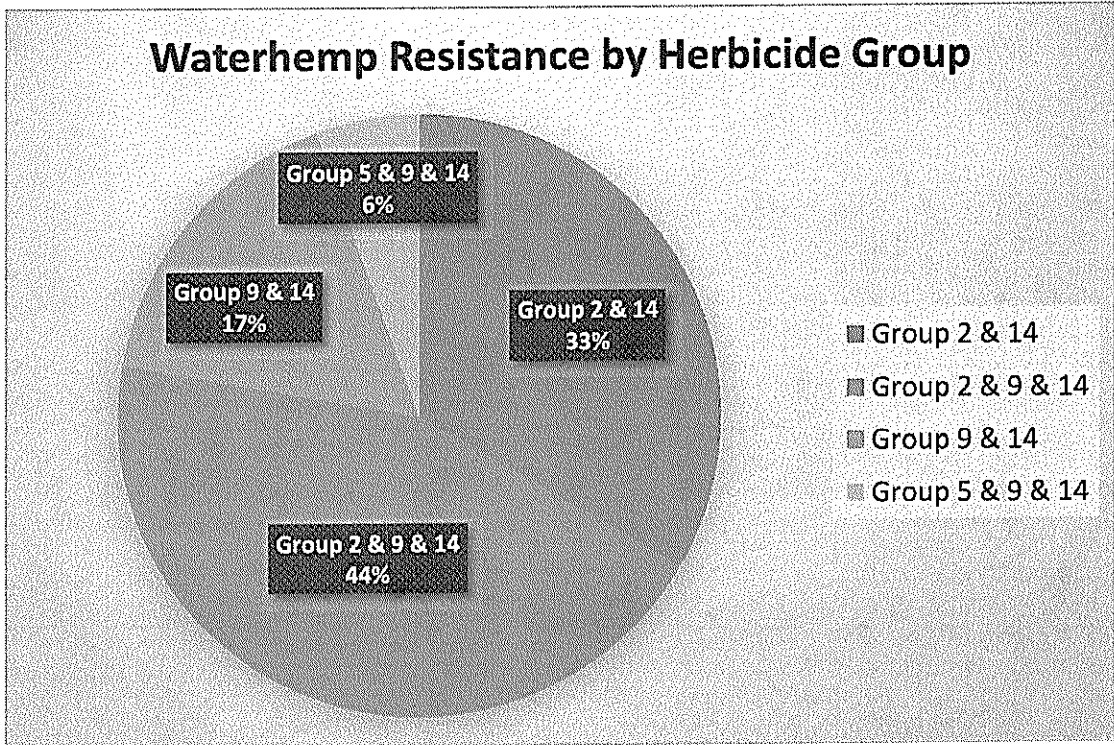
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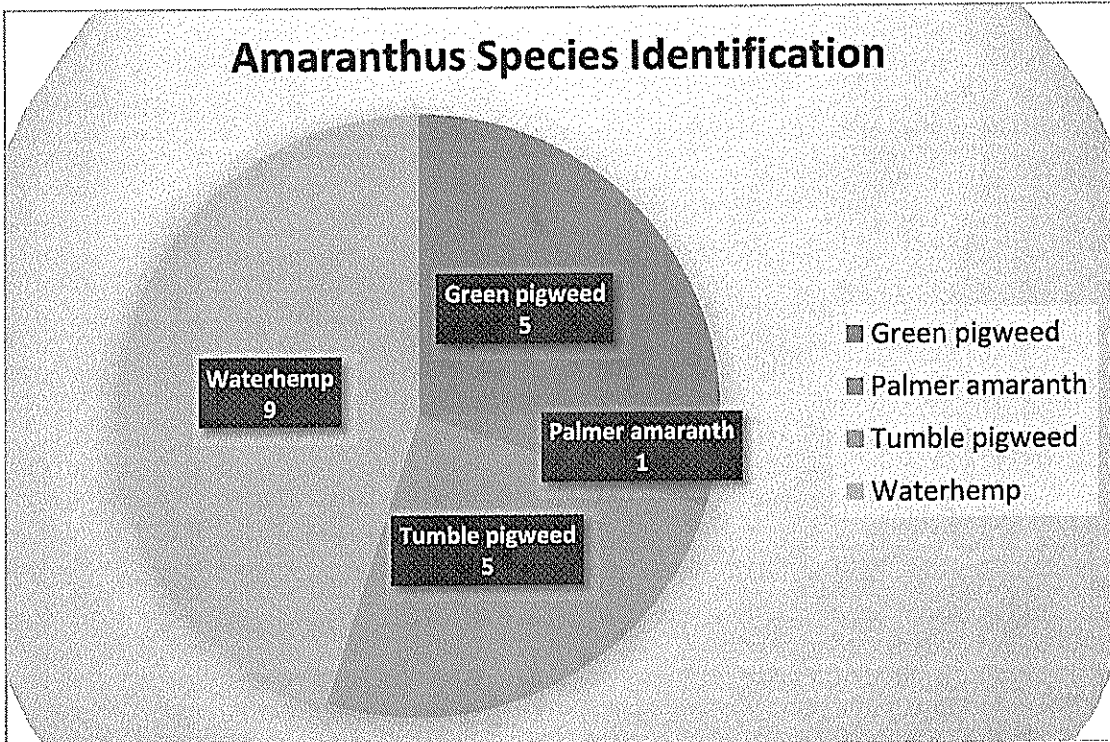
Total Number of Fields = 18



Total Number of Fields = 18



Total Number of Fields = 18



Total Number of Fields = 20