



Tissue Culture-Based on Virus Elimination in Stone Fruits: Method and Success Rate

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Tissue culture is an essential method in virus elimination for stone fruits, providing a reliable and effective technique to ensure the health and productivity of these valuable crops. This method employs meristem culture combined with thermotherapy or chemotherapy to produce virus-free plants. The process focuses on isolating the smallest, virus-free meristematic tissues, which are less likely to carry viral infections. By culturing these tissues in sterile, nutrient-rich environments, healthy plantlets can be developed, which can be used for large-scale propagation. The success rate of virus elimination through tissue culture in stone fruits, including cherries, peaches, and plums, varies depending on the type of virus, plant species, and method applied. Typically, success rates range from 70-95%, with higher efficacy reported when combining meristem culture with advanced viral detection techniques. Overall, tissue culture offers a high-potential approach for virus control, enhancing the quality, yield, and lifespan of stone fruit crops.

Keywords: Tissue culture, stone fruits, success rate, plant regeneration, virus-free plants, shoot tip culture, micropropagation, meristem culture, in-vitro propagation, cryotherapy, thermotherapy, virus elimination

Introduction to virus elimination in stone fruits: Tissue culture is widely used for virus elimination in stone fruits like cherries, plums, peaches, and apricots. This technique involves growing plant cells or tissues in a sterile environment, allowing for controlled conditions that can eliminate viruses and pathogens. By culturing meristematic tissues, often combined with heat or chemotherapy treatments, it is possible to produce virus-free plantlets. Success rates for virus elimination in stone fruits vary, often ranging from 70% to 90%, depending on factors like plant species, virus type, and the method's precision. This method plays a critical role in producing healthy planting materials, enhancing crop yields and quality.

Understanding tissue culture in plant propagation: Tissue culture is widely used for virus elimination in stone fruits like cherries, plums, peaches, and apricots. This technique involves growing plant cells or tissues in a sterile environment, allowing for controlled conditions that can eliminate viruses and pathogens. By culturing meristematic tissues, often combined with heat or chemotherapy treatments, it is possible to produce virus-free plantlets. Success rates for virus elimination in stone fruits vary, often ranging from 70% to 90%, depending on factors like plant species, virus type, and the method's precision. This method plays a critical role in producing healthy planting materials, enhancing crop yields and quality.

Methods of Virus Elimination in Stone Fruits Using Tissue Culture: Tissue culture methods are widely used for virus elimination in stone fruits due to their ability to produce virus-free plants from infected stock. One of the most effective techniques is meristem

culture, which involves isolating the meristem—a tiny, rapidly dividing region at the tip of a plant shoot—because viruses often do not infiltrate this part. By excising and culturing this meristematic tissue under sterile conditions, virus-free plants can be propagated even if the original plant was infected. Other complementary methods include thermotherapy and cryotherapy. These methods allow for the large-scale production of virus-free stone fruit plants, which are crucial for ensuring healthy, productive orchards and high-quality fruit yields.

- 1. Meristem culture:** Meristem culture is one of the most effective methods for virus elimination, relying on the fact that viruses often fail to reach the meristem—a tiny, actively dividing tissue at the shoot or root tips of plants. This region is less likely to be infected due to the plant's fast cell division in the meristematic region, which can outrun viral spread. In meristem culture, a small piece of meristematic tissue, usually only 0.1–0.3 mm in size, is carefully excised and placed on a nutrient-rich medium under sterile conditions to promote growth. The process involves careful control of nutrients, light, temperature, and humidity, allowing this tiny, virus-free tissue to grow into a full, healthy plantlet. With proper handling, meristem culture has a high success rate in generating virus-free plants, particularly in stone fruits, which are susceptible to various viruses impacting yield and quality.
- 2. Thermotherapy:** Thermotherapy complements meristem culture by using controlled heat to reduce viral activity within the plant. The plant tissue is subjected to elevated temperatures (usually between 35–40°C) over days or weeks. This heat stress can suppress or inactivate the virus, weakening it enough to prevent it from spreading to the meristematic tissue, which is then used for culture. For instance, thermotherapy might be applied to a whole plant or just parts of it, and afterward, meristematic tissue is isolated and cultured. Since different viruses have varied heat tolerances, thermotherapy protocols must be carefully tailored. The combination of thermotherapy with meristem culture has been shown to increase the overall efficiency of virus elimination, although not all plants tolerate prolonged heat exposure well, so the duration and intensity of treatment must be optimized based on the plant and virus type.
- 3. Cryotherapy:** Cryotherapy is a more recent approach, where plant tissues, often shoot tips, are exposed to extremely low temperatures (often using liquid nitrogen) to freeze and kill viral cells selectively, while sparing the resilient meristematic cells. In this method, shoot tips are subjected to a cryoprotectant solution to prevent cellular damage, then rapidly frozen. When thawed, the surviving cells, typically from the meristem, are isolated and cultured. Cryotherapy's benefit lies in its high precision, allowing it to eliminate even persistent viruses that may resist thermotherapy or standard meristem culture. However, the equipment and expertise required for cryotherapy are more specialized, so it is less commonly used outside advanced laboratories. Nevertheless, cryotherapy is particularly effective for viruses that are challenging to eliminate through other methods.
- 4. Combined applications and success rates:** The combination of these techniques can significantly increase the success rate of virus elimination, particularly for complex or resistant viruses. For instance, using thermotherapy before meristem culture or cryotherapy ensures that viral activity is minimized, allowing for healthier initial tissue samples. Success rates of these methods depend on factors like the specific virus, the type of stone fruit species, and conditions of the tissue culture environment. While meristem culture alone has a success rate between 60–80% for many viruses, the addition of thermotherapy or cryotherapy can push these rates even higher, approaching nearly 90% or more in some cases.

Role of Meristem Culture in Virus-Free Stone Fruit Production: Meristem culture plays a pivotal role in the production of virus-free stone fruits by leveraging the unique characteristics of the meristematic tissue, which is typically free of viruses due to its undifferentiated nature. By excising the meristem tips and culturing them in sterile conditions, plant breeders can generate healthy plantlets that are free from viral infections. This technique not only ensures the propagation of disease-resistant varieties but also enhances the overall productivity and longevity of stone fruit crops. Additionally, meristem culture facilitates rapid multiplication, allowing for the swift establishment of virus-free planting material, which is essential for sustainable fruit production.

Success Rate and Factors Influencing Virus Elimination: The success rate of virus elimination in stone fruits through tissue culture techniques—primarily meristem culture, thermotherapy, and cryotherapy—depends on factors like the type of virus, plant species, and conditions of the culture environment. Meristem culture alone can achieve a success rate of around 60–80%, as the meristematic region is often virus-free. However, combining it with thermotherapy, which uses controlled heat to suppress viruses, or cryotherapy, which freezes plant tissue to kill viral cells selectively, can boost success rates to nearly 90% or higher. The effectiveness of each method also varies depending on how well the plant tolerates temperature treatments and the specific virus involved, making precise protocol adjustments essential for optimal outcomes.

Case Studies and Examples in Stone Fruit Tissue Culture: Case studies in stone fruit tissue culture illustrate the effectiveness of virus elimination techniques. For instance, research on cherry and peach varieties has shown that applying meristem culture combined with thermotherapy can lead to successful propagation of virus-free plants, significantly improving yields and fruit quality. In one study, over 90% of the regenerated peach plants were free from the prevalent Peach Mosaic Virus, demonstrating the high efficiency of these methods. Similarly, experiments with plum trees indicated that cryotherapy effectively removed resistant viruses, further enhancing the success rates of tissue culture. These examples underscore the potential of tissue culture techniques to produce healthy, productive stone fruit varieties while mitigating the impact of viral diseases.

Challenges and Limitations of Tissue Culture in Virus Elimination: Despite the advantages of tissue culture for virus elimination in stone fruits, several challenges and limitations persist. One significant challenge is the risk of contamination during the culturing process, which can compromise the integrity of the tissue cultures and lead to the introduction of new pathogens. Additionally, not all viruses are effectively eradicated by tissue culture methods, particularly those that may reside in tissues not included in the culture, resulting in potential re-infection. The success rates can also vary significantly among different plant species and virus types, necessitating tailored approaches that require extensive research and optimization. Moreover, the resources and expertise needed for tissue culture can limit its accessibility for some growers, making it less practical for small-scale operations. Lastly, there can be high costs associated with setting up and maintaining tissue culture facilities, which may pose barriers to widespread adoption.

Future Directions and Advancements in Virus Elimination for Stone Fruits: Future advancements in virus elimination for stone fruits will likely focus on integrating cutting-edge biotechnological approaches with traditional tissue culture methods. Emerging techniques such as CRISPR gene editing hold promise for developing virus-resistant plant varieties by targeting specific viral genes, potentially reducing reliance on tissue culture for virus eradication. Additionally, advances in molecular diagnostics will enhance the detection of viruses at earlier stages, allowing for more precise and timely interventions. Exploring the

use of biopesticides and natural plant extracts could offer alternative strategies for managing viral infections in orchards. Combining these innovative methods with established tissue culture practices may significantly improve the efficiency and success rates of producing healthy, virus-free stone fruit plants, ultimately supporting sustainable agriculture and increasing crop resilience.

Conclusion

Tissue culture has emerged as an effective method for virus elimination in stone fruits, such as peaches and cherries. By utilizing techniques like meristem culture and thermotherapy, plant tissues are cultivated under sterile conditions to produce virus-free plantlets. The success rate of these methods varies, but many studies report up to 90% efficiency in eliminating specific viruses. This approach not only enhances the health and yield of stone fruit crops but also plays a crucial role in the propagation of disease-resistant varieties, contributing to sustainable agricultural practices.

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