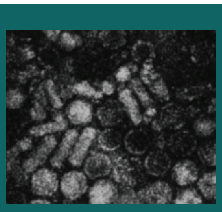

MUSHROOM PEST & DISEASE

MU16003 **fact sheet #8**

Mushroom Virus X Syndrome

Patch Disease and Brown Cap Mushroom Disease

GROWERS' SUMMARY

- Mushroom Virus X syndrome is a viral condition where two different diseases, Patch Disease and Brown Cap Mushroom Disease express.
- The viruses causing MVX syndrome in Australia have not been fully identified but are suspected to be different to those causing the disease in the United Kingdom and Europe.
- Don't ignore sporadic 'off white' or 'brown' mushrooms appearing – it might be an early warning that virus is replicating on your farm.
- MVX syndrome symptoms may appear similar to symptoms expressed by other diseases.
- If you suspect a virus infection, send samples for testing immediately.
- MVX syndrome viruses can be spread by mushroom spores and mycelium.
- Consider reducing the proportion of open mushrooms grown to reduce the number of mushroom spores on the farm.
- Reduce dust levels around the farm.
- Protect vulnerable operations – Phase II, Phase III and casing – from dust.
- Stringent hygiene based on efficient cook out and effective cleaning is the most successful management tool.

1. INTRODUCTION

Mushroom Virus X syndrome (MVX) describes a range of symptoms including delayed opening, reduced yields, distorted mushrooms and cap browning which express on cultivated *Agaricus bisporus* crops. MVX syndrome is now known to be a combination of two separate virus diseases – Patch Disease and Brown Cap Mushroom Disease – which can occur together and express a variable and complex symptomology. In some cases, one or two symptoms may predominate.

MVX syndrome first emerged in Britain in 1990 when mushroom crops began expressing symptoms resembling a virus infection. But established techniques used previously to detect the La France virus were unable to detect a virus responsible for the novel infection, leading researchers to investigate other causes.

It wasn't until a newly developed methodology was applied to samples from symptomatic farms that novel dsRNA bands were observed. The causal agent was found to be a suite of viruses which were identified by comparing the dsRNA bands isolated from samples from both disease-affected farms and high-yielding symptom-free farms.

Particularly vulnerable to infection are farm operations where compost is exposed, especially Phase II, Phase III and casing. Indeed, MVX syndrome appears particularly adapted to Phase III bulk handling facilities, but all farms are susceptible to the syndrome at one point or another.

MVX syndrome has been recorded from a number of mushroom growing areas including the United Kingdom and mainland Europe and most recently, Australia.

The little information available on the cost of the disease indicates the financial impact is significant, totalling some £50m in the UK during the year 2000 alone, impacting 80% of commercial growers and resulting in farm closures and the loss of up to 800 jobs.

2. SYMPTOMOLOGY

Like La France disease, MVX syndrome can be difficult to recognise in the early stages of infection. When there are low levels of virus on the farm, the only symptom expressing on a white strain may be the appearance of an occasional brown mushroom which is often put down to being 'one of those things' and ignored. But the longer the disease goes unrecognised the more the virus replicates and the amount of virus on the farm increases.

Once levels of MVX syndrome viruses exceed the farm's disease threshold, symptoms typical of virus infection will express. Because they often occur together, the symptoms of Patch Disease and Brown Cap Mushroom Disease may express on the same bed and appear as a single infection.

Video

Click on the link above to view a short video describing virus symptomology

Symptoms expressed in Patch Disease (Fig. 1, Fig. 2a)

- bare patches of unproductive compost which 'spiral' along the bed
- mushrooms have poor shelf-life

Figure 1 MVX symptomology. Patches of unproductive bare casing 'spiralling' along an affected bed. *Image: farm supplied*

- mushrooms have overall poor quality
- early opening
- crop delay
- yield reduction
- distorted mushrooms

Symptoms expressed in Brown Cap Mushroom Disease (Fig. 2b)

- mushroom caps become brown or white strains become 'off white' or 'cream'
- mushrooms have poor shelf life
- mushrooms have overall poor quality
- mushrooms tend to open prematurely

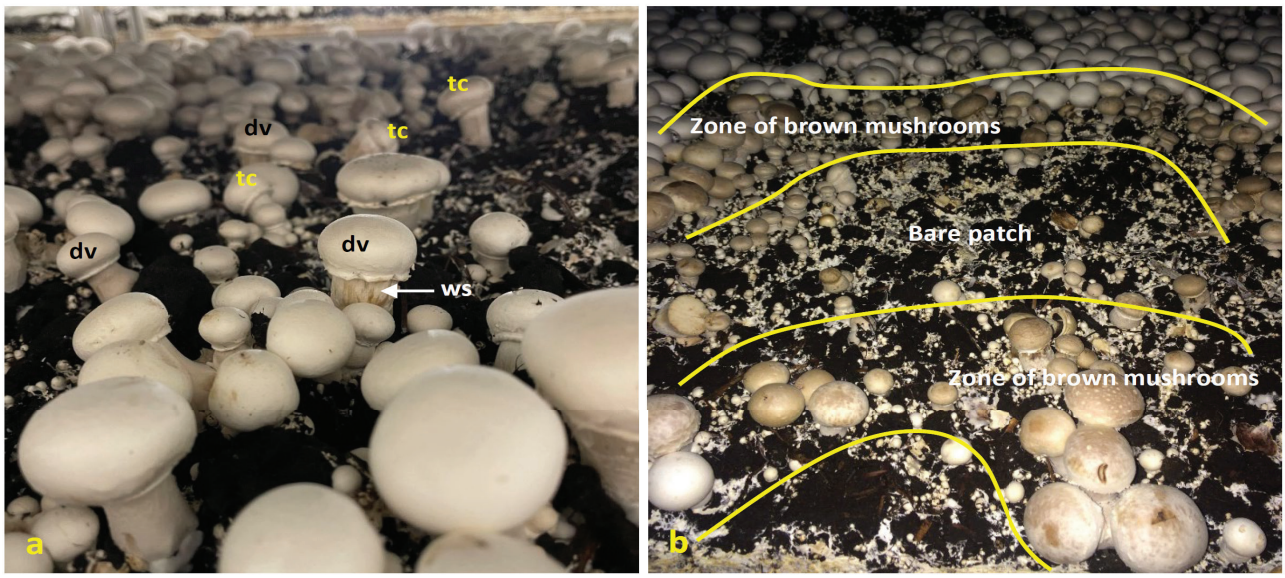

Figure 2 MVX symptomology. **a)** distorted mushrooms on an affected bed include dropped veils (dv), tilted caps (tc) and watery streaks (ws) **b)** brown, poor quality mushrooms expressing Brown Cap Mushroom Disease symptoms around the edge of a bare patch of casing. *Images: farm supplied*

3. MVX VIRUSES

Mycoviruses

The first ever mycoviruses – viruses that live inside fungal hyphae – were discovered in cultivated *A. bisporus* while investigating the cause of what was to become known as La France disease in the middle of last century. Although now known to occur widely throughout the fungal kingdom, most mycoviruses are benign and do not cause a reaction from their host. However, a small number do cause significant disease in *A. bisporus* crops.

Virus diseases of cultivated *A. bisporus* are caused by at least two different virus particles present in a synergistic relationship and the relative ratios of each virus may influence both the symptoms expressed and the degree of expression.

For La France disease, both La France Isoform Virus (LIV) and Mushroom Bacilliform Virus (MBV) have been isolated from affected mushrooms in both the UK and Australia. But for the MVX syndrome complex, the causal viruses appear to be different (Table 1) despite the same symptoms expressing.

Table 1 Viruses associated with *Agaricus* diseases

DISEASE	UK	AUSTRALIA
La France	LIV MBV	LIV MBV
Patch	AbV6 AbEV1*	AbV6 MBV
MBCD	AbV6 AbV16	AbV6

Patch Disease

Patch Disease, more prevalent on mainland Europe than the UK, was found to be associated with two viruses, Agaricus bisporus Virus particle 6 (AbV6) and Agaricus bisporus Edornavirus particle 1 (AbEV1). The role of AbEV1 in Patch Disease has not been confirmed.

In Australia, AbV6 and MBV have been detected in samples from symptomatic crops. Because routine mushroom virus sampling did not include AbEV1, the status of this virus is unknown. The significance of MBV detected in Australian Patch Disease is also unknown.

Brown Cap Mushroom Disease

In the United Kingdom where Brown Cap Mushroom Disease is more prevalent, two different viruses – AbV16 (comprising two distinct RNA molecules, dsRNA1 and dsRNA2) and AbV6 – have been isolated from affected crops. But in Australia, only AbV6 has been detected in mushrooms expressing Brown Cap Mushroom Disease symptoms. AbV16, also known as the Brown Cap Mushroom Virus (BCMV) has not been detected.

The Australian situation

The viruses detected in Australian mushroom crops expressing Patch Disease and Brown Cap Mushroom Disease (MVX syndrome) symptoms apparently differ to those detected in the UK and Europe. For Patch Disease, this is not so concerning as AbEV1 is not routinely tested for in Australia and the role of the virus in European disease expression is uncertain.

Up until the end of 2021, tests on a small number of symptomatic samples have failed to detect AbV16. The absence of AbV16 here in mushrooms expressing Brown Cap Mushroom Disease symptoms is significant and indicates either a novel and as yet unidentified virus is present in Australia or that Patch Disease is expressing a different symptomology to that in Europe.

4. DISEASE DEVELOPMENT

Crop infection

The mushroom crop is most vulnerable to MVX syndrome infection whenever the compost is exposed.

Infection often occurs when contaminated compost is introduced into Phase III compost during tunnel emptying and room filling. Contaminated filling heads, conveyors and rufflers spread tiny points of inoculum throughout the entire room during operations, leading to a heavily infected crop.

Likewise, dust contamination at spawning, during spawn-run and at casing also results in heavy infection. Very small virus-infected mycelial fragments generated during handling of fully run compost and infected mushroom spores can become airborne or incorporated into the farm 'dust'.

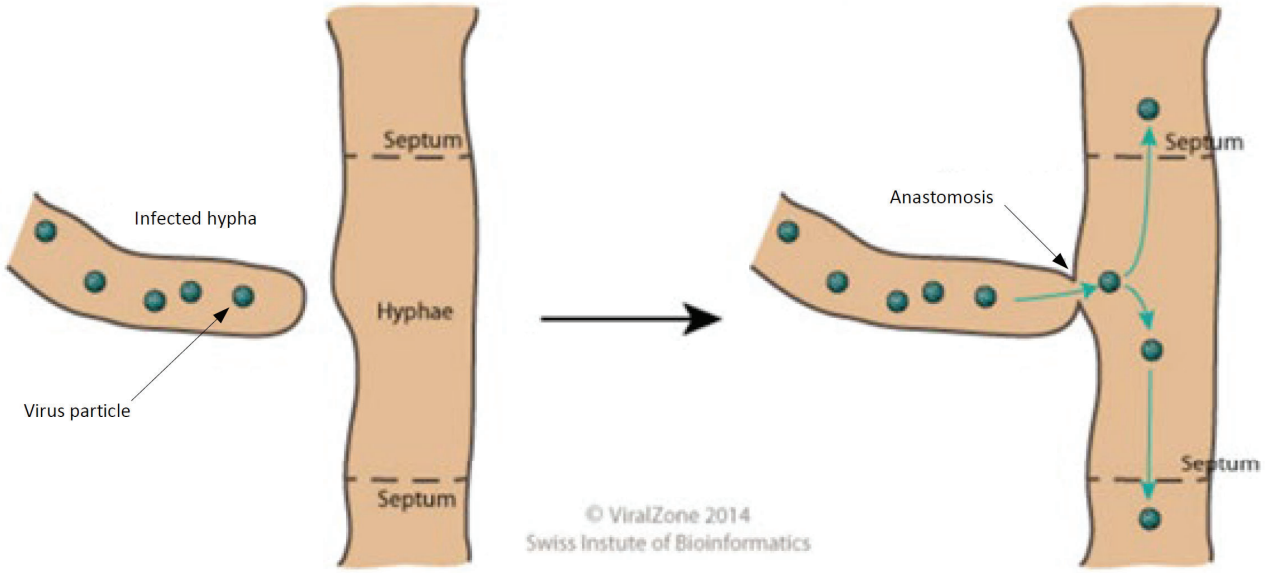

Figure 3 Hyphal anastomosis in fungal mycelium. *Image:* adapted from ViralZone 2014, Swiss Institute of Bioinformatics

Horizontal transmission

Once infected organic material is introduced into Phase III compost, contaminated mushroom hyphae begin to grow and branch, radiating out to colonise the compost. At the same time, the *Agaricus* mycelium, recovering from emptying and filling, also begins to grow and re-establish mycelial connections.

As one hypha encounters another, they touch and fuse in a process called anastomosis (Fig. 3) which forms a complex mycelial network. Nutrients, water, oxygen and some cell components then move from hypha to hypha to sustain the developing mycelium.

At the same time, virus particles are also transported from infected hyphae into uninfected hyphae and throughout the developing mycelium. The spread of virus particles by anastomosis is called horizontal transmission and it is the only way virus particles can spread within a crop of the same generation.

Disturbance of infected casing, compost and mushrooms by standard cultivation practices creates airborne mycelial fragments which are capable of germinating and initiating new infections either as separate fragments or as a component of farm dust.

Vertical transmission

As mushrooms begin to develop and differentiate, the hyphae containing the virus particles become incorporated into the fruitbody and viruses pass from the cap hyphae into the spores (Fig. 4). Because infected spores will initiate the *next* generation of infection, this process is known as vertical transmission.

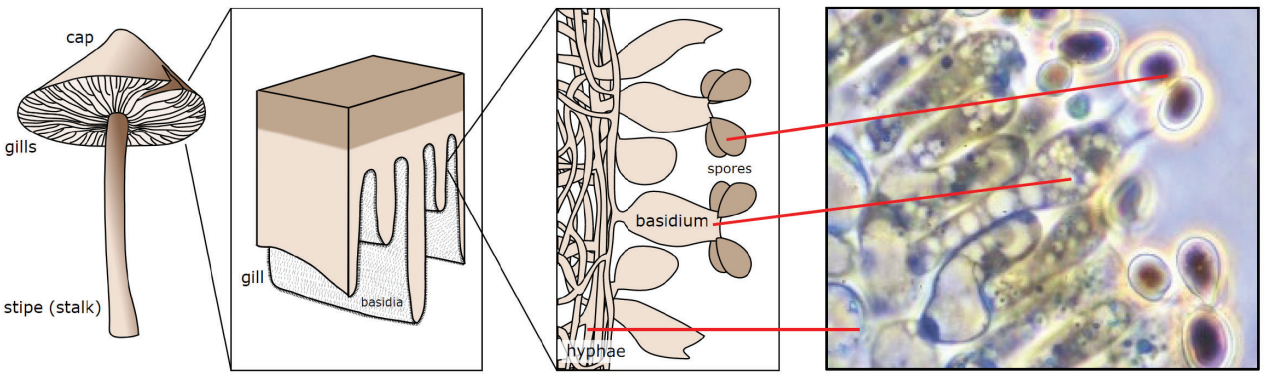

Figure 4 Spore development in basidiomycetes. The dispersal of virus-infected spores is called vertical transmission. *Line diagrams:* Debivort / CC BY-SA (http://creativecommons.org/licenses/by-sa/3.0/) | *Photograph:* Warwick Gill

Because the of the high infection rate and the huge numbers of spores produced – an 8cm diameter mushroom will produce over 1.3 billion spores at 40 million spores per hour – transmission within spores is a significant source of virus infection on-farm.

Spores can become airborne and start new infections wherever they settle and germinate, or they may become part of the farm dust.

5. MANAGEMENT STRATEGIES

A key action checklist has been provided (Table 2) to help assist with MVX syndrome management on your farm.

Because MVX syndrome viruses can only exist and be transmitted within infected host mushroom mycelium and spores, stringent hygiene is the most effective disease management tool. In addition to an efficient cook out, it is crucial that cleaning is done properly and effectively.

Most important are the initial cleaning steps. Because disinfectants are unable to penetrate compost to kill the infected mushroom mycelium and the spores inside, the compost and other organic material must be physically removed from a surface prior to sanitizing. Once disinfectants are applied to the cleaned surfaces, the 'time to kill' is critical – disinfectants must remain wet on the surface for the required time as per manufacturer's instructions.

Dust management is also an important aspect of virus disease control. Reduce the amount of dust on the farm and protect vulnerable Phase II, Phase III and casing operations from dust contamination.

If a virus is at all suspected, send samples for testing. In the early stages, MXV syndrome symptoms may appear similar to other diseases such as nematode infestation and Mummy Disease. Eliminating a virus as the cause of expressed symptoms allows the farm to focus more effectively on developing better crop management and hygiene systems. While waiting for test results, review farm hygiene and farm vulnerabilities based on the biology of the organism and the method of spread.

6. KEY REFERENCES

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Table 2 Checklist of key action points for prevention and control of MVX syndrome

Location	✓	×	?	Action point
Phase II and III	☑	☒	☐	HEPA filters installed on Phase II and Phase III air intakes – avoid entry of unfiltered air
	☑	☒	☐	All filters and ducts regularly maintained
	☑	☒	☐	Phase II spawning halls completely separated from Phase III emptying areas
	☑	☒	☐	Phase III emptying and spawning are not carried out at the same time
	☑	☒	☐	Phase III emptying halls maintained under neutral pressure
	☑	☒	☐	Spawning halls maintained under positive pressure with filtered air
	☑	☒	☐	Spawn cool room storage racks and transport freight pallets cleaned and disinfected
	☑	☒	☐	Spawn stored in dedicated cool room
	☑	☒	☐	Partially used spawn bags **not** returned to cool room
	☑	☒	☐	Spawning equipment cleaned and disinfected immediately before and after use
	☑	☒	☐	Machinery for handling Phase II and Phase III compost is not shared
	☑	☒	☐	Phase III tunnels, spawn-run shelves and spawn-run trays are cleaned and steamed before re-use
	☑	☒	☐	Phase III tunnel nets cleaned and disinfected
	☑	☒	☐	Phase II and Phase III tunnels rotated on a regular basis between compost batches
Filling & casing	☑	☒	☐	Casing material is stored under cover
	☑	☒	☐	All dust removed from the outside of peat bags before opening
	☑	☒	☐	Casing mixing area sanitized before and after use
	☑	☒	☐	Casing performed under positive pressure to prevent contamination by spores in airborne dust
	☑	☒	☐	Only laboratory-prepared casing inoculum used if farm is known to have virus
	☑	☒	☐	Adjacent concrete areas are dampened down during casing and filling to reduce airborne dust
	☑	☒	☐	All equipment used for casing and filling effectively cleaned and disinfected before and after use
	☑	☒	☐	All casing and filling staff wear fresh clean clothes before operations begin
	☑	☒	☐	During operations, casing and filling staff do not share facilities with harvest and post-harvest staff
	☑	☒	☐	Grow room doors kept closed during filling; do not harvest, clean or empty rooms during filling
	☑	☒	☐	Concrete in front of grow room sanitised immediately before and after fill
	☑	☒	☐	Diseased grow rooms kept closed, no grow rooms emptied during casing and filling
	☑	☒	☐	Excess Phase III compost removed from site immediately, not stockpiled
Grow room	☑	☒	☐	Pick regularly to prevent affected mushrooms opening and dispersing virus in infected spores
	☑	☒	☐	Reduced cropping of open mushrooms considered
	☑	☒	☐	Waste mushroom material, butts and stalks disposed of regularly offsite
	☑	☒	☐	Grow room exhaust air filtered if growing open mushrooms
	☑	☒	☐	Concrete apron and/or corridor floor outside affected grow room disinfected daily
	☑	☒	☐	Grow room floors, shelves and walls washed and disinfected after emptying
	☑	☒	☐	Concrete apron outside the grow room washed and disinfected immediately after emptying
	☑	☒	☐	Netting cleaned and disinfected after emptying; if MVX present on farm, re-steam
	☑	☒	☐	Dust kept to a minimum reducing chances of airborne contamination – no dry sweeping after harvest
Crop termination	☑	☒	☐	Crop cooked out *in situ* at 65–70°C for 12 hours
	☑	☒	☐	Temperature probes calibrated regularly
	☑	☒	☐	Grow room floor, particularly cracks and joins, is sanitized after cookout
	☑	☒	☐	Steam introduced gradually to prevent sudden in-rush of air, dispersing *Agaricus* spores
	☑	☒	☐	Cookout room sealed tightly including drains to prevent spore release during steam introduction
	☑	☒	☐	Ensure crop is well covered with an approved disinfectant before emptying if cookout not possible
	☑	☒	☐	Early crop termination considered especially if there is heavy infection
	☑	☒	☐	Spent mushroom compost removed immediately after cookout, not stockpiled on site
Monitoring	☑	☒	☐	Viral levels monitored at regular intervals by sending mushroom samples for diagnosis
General	☑	☒	☐	All returnable containers are sanitized before taking into grow room
	☑	☒	☐	Transport vehicles and equipment moving between farms are cleaned and disinfected regularly
	☑	☒	☐	Farm dust is well managed
	☑	☒	☐	Buildings, roadways and all concrete areas disinfected daily
	☑	☒	☐	Farm maintenance up to date; rooms and smooth surfaces in good condition are easier to keep clean
	☑	☒	☐	Avoid the accumulation of organic matter around the farm which can act as source of infection
	☑	☒	☐	General farm sanitation maintained at a very high standard

Correlated from Fletcher & Gaze (2008) and Archibald-Fleming et al *(2015)*

MU16003 - Pest and Disease Management and Research Services

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