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AAT & ALAT ACTIVITY IN *CATLA catla* EXPOSED TO PYRACLOSTROBIN 20% (WG)

Abubakkar.V **, Aswartha Narayana. A **, Mounika. G ** and V.VenkataRathnamma*

**Research Scholars Dept.of.Zoology & Aquaculture

*Professor Dept.of.Zoology & Aquaculture

**/*Acharya Nagarjuna University, Nagrjuna Nagar, Guntur-522510

ABSTRACT

The changes in the levels of AAT and ALAT were studied in different tissue of brain, muscle, gill, liver and kidney in the test fish *Catla catla* under sub-lethal and lethal concentrations of Pyraclostrobin (20% WG) after 24h depletion was in liver (35.46), followed by gill (19.32), brain (23.1), kidney (15.89), muscle (14.31). Under lethal exposure to Pyraclostrobin for 24h, the percentage of AAT depletion in liver (37.23), brain (24.11), gill (20.46), kidney (16.78) and in muscle (15.12). Under sub-lethal exposure to Pyraclostrobin for 4days the percentage of AAT depletion was liver (33.45) brain (21.7), gill(18.11), kidney(14.21) and muscle(12.17). Under sub-lethal exposure to Pyraclostrobin for 8day of AAT depletion was in liver(32.34), brain(20.09), gill(17.97), kidney(13.26) and muscle(11.25). Under sub-lethal concentrations for 24h of Pyraclostrobin (20% WG), in ALAT activity in *Catla catla* is in the order of: 24 h depletion was in liver (17.02), gill (18.32), brain (22.42), kidney (14.89), and muscle (20.41). Under sub-lethal exposure to Pyraclostrobin for 4days ALAT in liver (16.53) brain (23.27), gill(17.16),(14.51) and muscle(20.12). Under sub-lethal exposure to Pyraclostrobin for 8day ALAT depletion was in liver(15.82), brain(22.34), gill(16.78), kidney(13.67) and muscle(19.18).

KEY WORDS: Pyraclostrobin , *Catla catla* AAT and ALAT and sublethal concentration

Introduction:

Acutely poisonous events most notably fish kills which were relatively mutual few decades ago are now rarely experiential in most industrialized countries; however, even sublethal toxicity could leads to plain impacts on entire populations (Richard *et al.*, 2012). Agriculture in Andhra Pradesh, India forms a main portion of the state economy. Large number of individuals of this state depends on agriculture provide with the

required food grains.

The production of paddy, chilies, tobacco, cotton, and other different kinds of crops are cultivated in wetland regions. Major part of cultivated lands is below Krishna River (Lat $15^{\circ} 18' - 16^{\circ} 50'$, long. $70^{\circ} 10' - 80^{\circ} 55'$ east) area, which include Krishna and Guntur Districts of Andhra Pradesh. In the current past the local agriculture officers advise the farmers to decrease indiscriminate pesticide scattering and abate the usage of banned insecticides. From the upland areas of this locality, the pesticides are washed to the low land water bodies through surface runoff, where the aquaculture actions are taken up by the farmers. Since both Krishna and Guntur Districts are maximum pesticides consuming places, the water is polluted by pesticides. It is important to know the impact of water qualities and the effect of pesticides to aquatic organisms, the more than a few pesticides us pyraclostrobin 20% (wg) on the non-target organisms are not so far investigation locally. Hence efforts have made to the toxicity of pyraclostrobin 20% (wg) to Indian major carp *Catla catla*.

Materials and Methods:

Fish *Catla catla* of size 6 ± 7 cm and 6.5 ± 2 g weight were brought from a local fish farm Kuchipudi, Guntur District of Andhra Pradesh, India and acclimatized at $28 \pm 2^{\circ}\text{C}$ in the laboratory for 15 days. Such acclimatized fish were exposed to sublethal and lethal concentrations of pyraclostrobin (20% WG) commercial grade for 24h, 4 and 8 days. The vital tissues like muscle, brain, liver, gill and kidney of the fish were taken for the estimation of Acetyl chloride esterase (AAT) and (ALAT) enzyme assays were performed spectrophotometrically by the method (Reitman and Frankel 1957)

AMINOTRANSFERASES :

In general, the pesticides increase the activities of some enzymes and decrease the activities of others; while the activities of a few enzymes remain unchanged in various tissues of horse mackerel (*Trachurus trachurus*) fish (Caglayan *et al.*, 2020). Aminotransaminases play an important role in the utilization of amino acids for the oxidation and gluconeogenesis (Kumar *et al.*, 2011; Zhang *et al.*, 2019). The AAT activity provides the oxaloacetate required for the gluconeogenesis pathway to meet the additional supply of glucose for the production of energy under reduced phase of oxidative metabolism (Rajkov *et al.*, 2018).

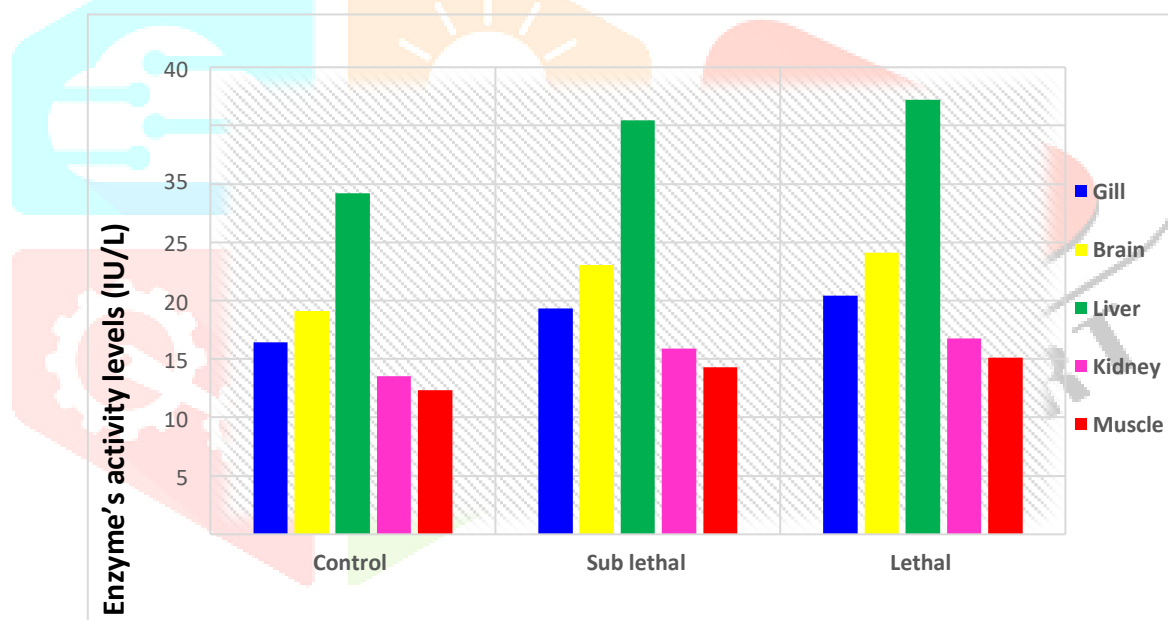
The ALAT activity measured in liver of eel tissues during exposure to propanol due the existence of heavy drain on metabolites during propels stress to provide inter mediates to the kerb's cycle

Changes in the specific activity levels of Aspartate aminotransferase (AAT) (μ moles of formazan /mg protein/hr) and percent change over control in different tissues of the freshwater fish, *Catla catla* exposed to sub-lethal and lethal concentrations of Pyraclostrobin (20% WG) for 24h.

Tissues	Control (mg/g)	Sub-lethal (mg/g)	% Change (mg/g)	Lethal(mg/g)	% Change (mg/g)
Gill	16.45 ±0.012	19.32 ±0.023	-17.44	20.46 ±0.032	-24.37
Brain	19.15 ±0.043	23.1 ±0.023	-20.62	24.11 ±0.086	-25.90
Liver	29.23 ±0.011	35.46 ±0.018	-21.31	37.23 ±0.094	-27.36
Kidney	13.56 ±0.024	15.89 ±0.014	-17.18	16.78 ±0.013	-23.74
Muscle	12.34 ±0.076	14.31 ±0.011	-15.96	15.12 ±0.076	-22.52

Values are the mean of five observations ;(±) indicates the standard deviation: Values are significantly at P< 0.05.

Changes in the specific activity levels of Aspartate aminotransferase (AAT) (μ moles of formazan /mg protein/hr) and percent change over control in different tissues of the freshwater fish, *Catla catla* exposed to sub-lethal and lethal concentrations of Pyraclostrobin (20% WG) for 24h.

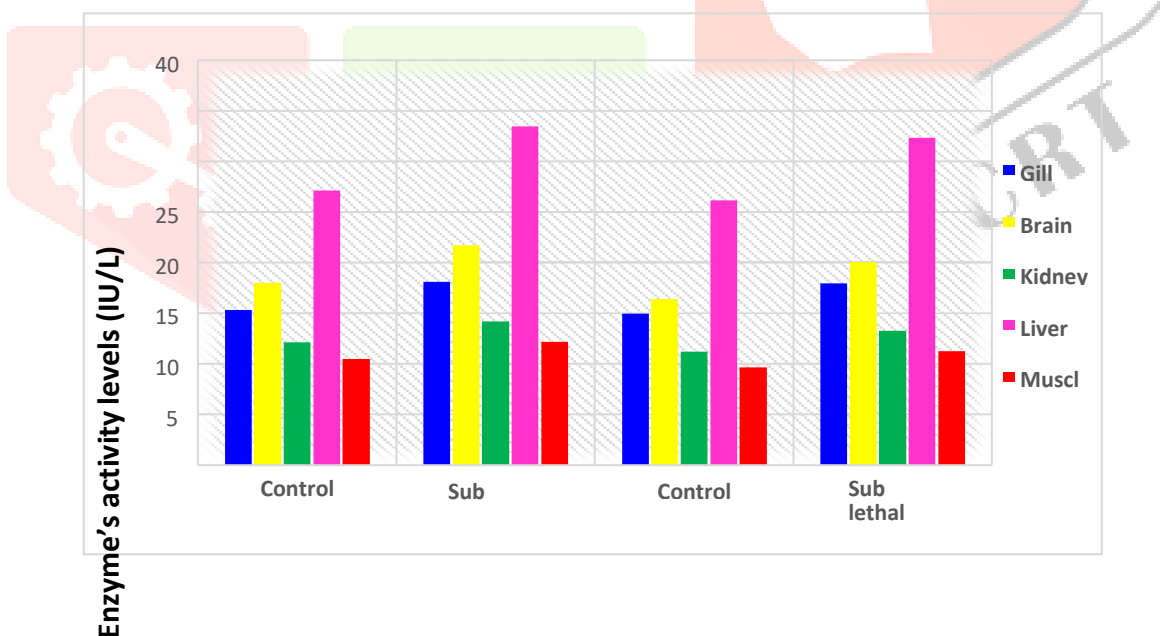


Changes in the specific activity levels of Aspartate aminotransferase (AAT)(μ moles of formazan /mg protein/hr) and percent change over control in different tissues of the freshwater fish, *Catla catla* exposed to sub-lethal concentrations of Pyraclostrobin (20% WG) for 4 and 8 days:

Tissues	4 Days			8 Days		
	Control (mg/g)	Sub-lethal (mg/g)	% Change (mg/g)	Control (mg/g)	Sub-lethal (mg/g)	% Change (mg/g)
Gill	15.33 ± 0.009	18.11 ± 0.043	-18.13	14.95 ± 0.044	17.97 ± 0.045	-20.20
Brain	17.98 ± 0.023	21.7 ± 0.034	-20.68	16.39 ± 0.087	20.09 ± 0.054	-22.57
Kidney	12.11 ± 0.011	14.21 ± 0.023	-17.34	11.23 ± 0.023	13.26 ± 0.033	-18.07
Liver	27.12 ± 0.035	33.45 ± 0.012	-23.34	26.12 ± 0.022	32.34 ± 0.033	-23.81
Muscle	10.47 ± 0.067	12.17 ± 0.022	-16.23	9.67 ± 0.042	11.25 ± 0.085	-16.33

Values are the mean of five observations ;(\pm) indicates the standard deviation: Values are significantly at $P < 0.05$.

Changes in the specific activity levels of Aspartate aminotransferases (AAT) (μ moles of formazan /mg protein/hr) and percent change over control in different tissues of the freshwater fish, *Catla catla* exposed to sub-lethal concentrations of Pyraclostrobin (20% WG) for 4 and 8 days.



Alanine Amino-Transferase (ALAT):

In general, the pesticides increase the activities of some enzymes and decrease the activities of others; while the activities of a few enzymes remain unchanged in various tissues of horse mackerel (*Trachurus trachurus*) fish (Caglayan *et al.*, 2020). Aminotransaminases play an important role in the utilization of amino acids for the oxidation and gluconeogenesis (Kumar *et al.*, 2011; Zhang *et al.*, 2019). The AAT activity

provides the oxaloacetate required for the gluconeogenesis pathway to meet the additional supply of glucose for the production of energy under reduced phase of oxidative metabolism (Rajkov *et al.*,2018). The ALAT activity measured in liver of eel tissues during exposure to propanol due the existence of heavy drain on metabolites during propolis stress to provide inter mediates to the kerb’s cycle (Sancho *et al.*, 2009; Ghanem *et al.*,2016). Similar increase in transaminase enzymes have been reported in fish tissues exposed to pesticides (Neelima *et al.*, 2016).

The pesticide stress was known to induce significant change in protein metabolism; it is likely that the amino transferases were also considerably affected. Increased activities of AAT and ALAT in different tissues of fish either increased operation of transamination or increased synthesis of amino acids from other sources like glucose of fatty acids during Pyraclostrobin (Ullah *et al.*, 2018). Present study Increase in aminotransferases activity was reported in fish *Catla catla* under Pyraclostrobin pesticides stress enhancement of activity of the transaminase provided the oxaloacetic acid and pyruvate α -ketoglutarate and glutaric acid to meet the increased energy demand during pesticide-imposed toxicity, increased levels of transaminases AAT and ALAT of fish.

Table.V.3.11.21. Changes in the specific activity levels of Alanine aminotransferases (ALAT) (μ moles of formazan /mg protein/hr) and percent change over control in different tissues of the freshwater fish, *Catla catla* exposed to sub-lethaland lethal concentrations of Pyraclostrobin (20% WG) for 24h.

Tissues	Control (mg/g)	Sub-lethal (mg/g)	% Change (mg/g)	Lethal (mg/g)	% Change (mg/g)
Gill	15.85 ± 0.011	18.32 ± 0.034	-15.58	19.56 ± 0.035	-23.40
Brain	19.15 ± 0.043	22.42 ± 0.056	-17.07	23.12 ± 0.033	20.73
Liver	14.23 ± 0.024	17.02 ± 0.065	-19.60	18.14 ± 0.024	-27.47
Kidney	13.56 ± 0.054	14.89 ± 0.044	-9.80	15.76 ± 0.026	-16.22
Muscle	17.67 ± 0.087	20.41 ± 0.035	-15.50	20.42 ± 0.018	-15.56

Values are the mean of five observations ;(\pm) indicates the standard deviation: Values are significantly at P< 0.05

Changes in the specific activity levels of Alanine aminotransferases (ALAT) (μ moles of formazan /mg protein/hr) and percent change over control in different tissues of the freshwater fish, *Catla catla* exposed to sub-lethaland lethal concentrations of Pyraclostrobin (20% WG) for 24h.

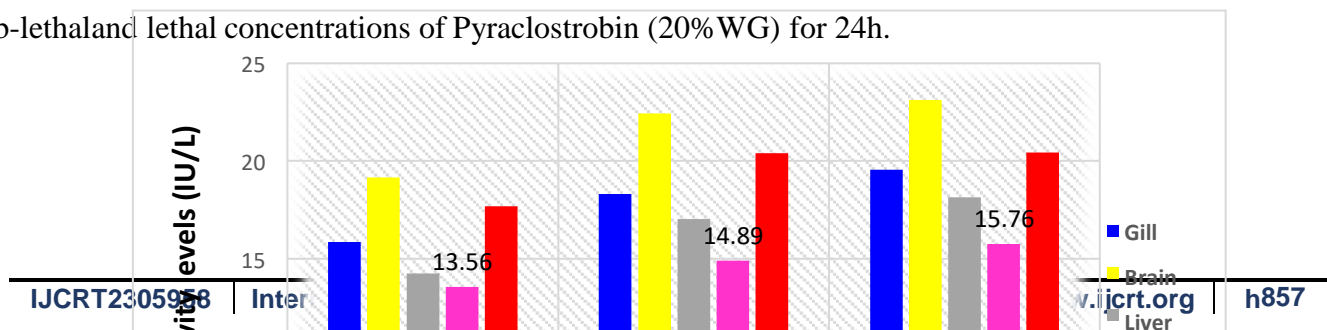
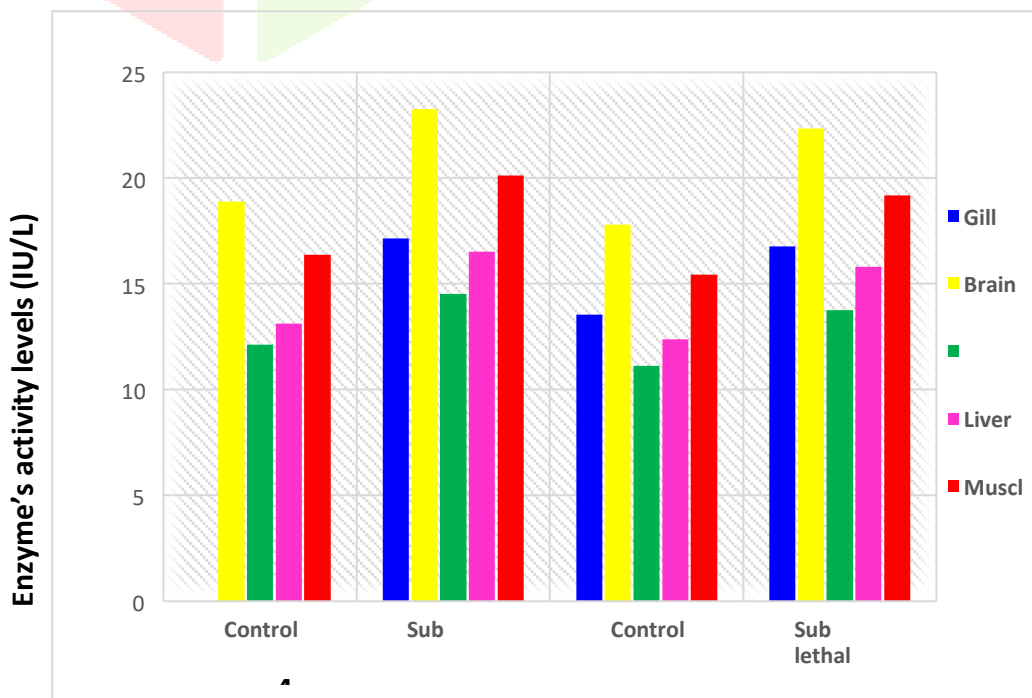


Table.V.3.11. 22.Changes in the specific activity levels of Alanine aminotransferases (ALAT) (μ moles of formazan /mg protein/hr) and percent change over control in different tissues of the freshwater fish, *Catla catla* exposed to sublethal concentrations of Pyraclostrobin (20%WG) for 4 and 8 days.

Tissues	4 Days			8 Days		
	Control (mg/g)	Sub-lethal (mg/g)	% Change (mg/g)	Control (mg/g)	Sub-lethal (mg/g)	% Change (mg/g)
Gill	14.27 ± 0.013	17.16 ± 0.045	-20.25	13.56 ± 0.054	16.78 ± 0.034	-23.74
Brain	18.89 ± 0.087	23.27 ± 0.076	-23.18	17.82 ± 0.034	22.34 ± 0.086	-25.36
Kidney	12.11 ± 0.045	14.51 ± 0.098	-19.81	11.13 ± 0.023	13.67 ± 0.098	-22.82
Liver	13.12 ± 0.098	16.53 ± 0.023	-25.99	12.37 ± 0.076	15.82 ± 0.044	-27.89
Muscle	16.38 ± 0.066	20.12 ± 0.012	-22.83	15.43 ± 0.097	19.18 ± 0.012	-24.30

Values are the mean of five observations ;(\pm) indicates the standard deviation: Values are significantly at $P < 0.05$

Changes in the specific activity levels of Alanine aminotransferases (ALAT) (μ moles of formazan /mg protein/hr) and percent change over control in different tissues of the freshwater fish, *Catla catla* exposed to sublethal concentrations of Pyraclostrobin (20% WG) for 4 and 8 days.



Increase in aminotransferases activity was reported in fish *Clarias batrachus*, under carbofuran pesticide stress, enhancement of activity of the transaminases provided the oxaloacetic acid and pyruvate, α -ketoglutarate and glutaric acid to meet the increased energy demand during carbofuran imposed toxicity, increased levels of transaminases AAT and ALAT of fish *C. Carpio* to the pesticide's glyphosate (Tazeen *et al.*, 2019). Significant increase in AAT and decrease in ALAT was observed in *Rhamdia quelen* exposed to sublethal concentration of cypermethrin., AAT and ALAT level increased in blood serum to treated with cypermethrin intoxicating fish *Cirrhinus mrigala* Since the pesticide stress was known to induce significant change in protein metabolism, it is likely that the amino transferases were also considerably affected (Canli *et al.*, 2018).

Increased activities of AAT and ALAT in different tissues of fish suggest either increased operation of transamination or increased synthesis of amino acids from other sources like glucose or fatty acids during thiodicarb intoxication. The increase in activities of aminotransferases as in the present study demonstrating a consistent increase in the activities of these enzymes under conditions of enhanced gluconeogenesis (Tazeen *et al.*, 2019; Raghu *et al.*, 2020). AAT and ALAT are located in both mitochondrial and cytosol fractions of the cell. A close relation appears to exist between the mitochondrial integrity and transaminase levels,(Konde *et al.*,2017). Tazeen *et al.*,(2019) modification in the organization of mitochondria is bound to alter the enzyme systems associated with it. De la Torre *et al.*,(2005) elevation in levels transaminases ALAT and AAT in fish *Cnesterodon decemmaculatus* using those enzymes as biomarkers of polluted water.

The significant increase in the activities AAT and ALAT in different tissues exposure of cypermethrin due to incorporation of ketoacids into the TCA cycle via generation of glutamate through tissue transamination followed by their conversion of α -ketoglutarate through oxidative deamination to favours gluconeogenesis or energy production (Prasanth and Neelagund.,2008; Ullah *et al.*,2018). Elevation of AAT and ALAT activities in eel *Anguilla Anguilla* under propanol intoxication (Sancho *et al.*,(2009). Increased ALT activity was noticed in *C. carpio* and *Oreochromis niloticus* exposed to cadmium (Ghelichpour *et al.*, 2020). AAT catalysis reversible transamination of glutamate and oxalo acetate to α -ketoglutarate and aspartate, while ALAT catalysis the reversible transamination of glutamate and pyruvate to α -keto glutamate and alanine (Nissen *et al.*,2017). Thus, the aminotransferases along with GDH contribute some strategic substances such as α -ketoglutarate, pyruvate, oxaloacetate, glutamate etc., to oxidative metabolism (Tazeen *et al.*, 2019).

The elevation of AAT activity provides the oxaloacetate require for the gluconeogenesis pathway to meet the additional supply of glucose for the production of energy under reduced phase of oxidative metabolism, increased activity of aminotransferases due to monostrophes in *Channa punctatus* (Raghu *et al.*,2020). Elevation in the levels of AAT in different tissues of brain, muscle, gill and kidney of the fish *Catla catla* can be ketoacids like α ketoglutarate and oxaloacetate for contributing to gluconeogenesis and or energy production

necessary to meet the energy demand under the toxic manifestations. The alterations in the levels of aminotransferases induce Pyraclostrobin clearly indicate that stress bring about the metabolic reorientation in the tissues by raising energy resources through transaminases systems. Ansari, S. and Ansari, B.A. (2014) glutathione (GSH) level decreased in zebrafish when exposed to dimethoate. Plasma membrane-derived enzymes which are associated with lysosomal activities, phagocytosis, digestion and transport of nutrients. Physiological changes in Freshwater fishes are widely used as a biomarker of any stressor component in the aquatic environment (Naglaa Elarabany, and Mohammed Bahnasawy.,2019). increasing lipid peroxidation in many organs such as brain, liver, kidney and gills (Nimmy, *et al.*, 2018).

CONCLUSION:

Present study Elevation in the levels of AAT and ALAT in different tissues of brain, muscle, gill and kidney of the fish *Catla catla* can be ketoacids like α ketoglutarate and oxaloacetate for contributing to gluconeogenesis and or energy production necessary to meet the energy demand under the toxic manifestations. The alterations in the levels of aminotransferases induce Pyraclostrobin clearly indicate that stress bring about the metabolic reorientation in the tissues by raising energy resources through transaminases systems

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