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钾对‘短枝’冬枣皱缩的影响

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摘 要：【目的】为明确钾与‘短枝’冬枣皱缩的关系及其生理机制，以便制定有效的防治方案。**【方法】**以‘短枝’冬枣为试验材料，研究不同时期(幼果期、膨大期、白熟期)叶面喷施 0.05 g·mL^{-1} K_2SO_4对果实皱缩率、细胞壁成分果胶和纤维素及其降解酶活性、果实硬度和显微结构的影响。**【结果】**不同时期喷钾均降低了果实皱缩率；不同程度地提高了果实果胶与纤维素含量，降低了果实果胶酶与纤维素酶的活性，增强了果实硬度，改善了果实显微结构。其中，果实膨大期喷钾对防止果实皱缩、增加果胶含量效果极显著，分别比对照降低60.65%和增加95.4%；膨大期和白熟期喷钾可保持果皮角质层的完整性，维持表皮细胞密实度和规整性，减少果肉空腔数量。又以膨大期喷施效果最好；果实皱缩率与果胶含量显著正相关，与果胶酶活性显著负相关，相关系数分别达0.997 6和0.994 6。果实皱缩率与纤维素含量及其分解酶活性的相关性不显著。**【结论】**果实膨大期叶面喷施K_2SO_4降低‘短枝’冬枣成熟期果实皱缩率效果最好。因此，7月20日起每隔7 d喷1次，共喷施3次 0.05 g·mL^{-1}的 K_2SO_4是防治‘短枝’冬枣成熟期果实皱缩的有效措施。

关键词：‘短枝’冬枣；皱缩；K_2SO_4；果胶；显微结构

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Effect of potassium fertilization on the shrinkage of ‘Short branch’ winter jujube

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Abstract:【Objective】Winter jujube is the most characteristic fruit in China. ‘Short branch’ winter jujube, also known as second generation of ‘Zhanhua’ winter jujube, is a new excellent bud mutation cultivar selected from the natural variation of winter jujube in Zhanhua district, Binzhou city, Shandong province. The shrinkage of ‘Short branch’ winter jujube initiates from the mature stage is a new disease occurring in Shandong, Hebei, Shanxi, Shaanxi and other major cultivation areas in recent years. Once the disease occurs, the fruit loses its commercial value. In 2019, the incidence in Zhanhua district and Wudi counties was 35.2%-41.0%. No pathogenic bacteria have been isolated from the shrunk part, and without symptoms such as flesh browning and bitterness. It is thus different from the traditional fruit shrink disease. The shrinkage has become an important factor restricting the development of winter jujube industry in China. Studies have shown that potassium is closely related to the metabolic process of fruit trees, and it is an activator of many enzymes, participating in the synthesis, transportation and transformation of sugars and starch. Potassium is effective to enhance the hydrophilicity of protoplast and cell osmotic pressure, increase the sugar reserve in the tree, increase the cellulose content in the epidermis of branches and fruits, and reduce the wrinkled fruit of citrus. The application of potassium fertilizer can also improve the shelf life, fruit firmness and coloration in peach fruit. Therefore, potassium is

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likely to be related to the shrinkage of 'Short branch' winter jujube. But the association between them is not clear. In order to verify their relationship and physiological mechanism, and then development an effective control method, this study was carried out.【Methods】The experiment was conducted in a winter jujube orchard in Zhanhua district and Wudi county, Binzhou city, Shandong province, from June to October 2018 and June to October 2019. According to the phenology of winter jujube, June 20 to July 20 was the young fruit stage, July 20 to August 22 the expansion stage, and August 22 the maturation stage of fruit. There were 4 treatments, the control, K_2SO_4 spray at young fruit stage, K_2SO_4 spray at expansion stage and K_2SO_4 spray at the early maturation stage. The concentration of the sprays was 0.05 g·mL^{-1}. For each stage, spray was conducted 3 times at an interval of 7 days around 18:00 pm until drip-off. Spraying water was used as the control. A single factor randomized experiment design was used. Each treatment had 3 repetitions each with 3 trees. There were 36 trees in total. Then protopectin and cellulose, activities of wall-degrading enzymes, cell wall component and microstructure and fruit hardness were analyzed.【Results】Spraying potassium at different stages reduced the rate of fruit shrinkage, increased the contents of pectin and cellulose in different degrees, decreased the activities of pectinase and cellulase, increased fruit hardness, and improved the microstructure of fruit. Among the treatments, spraying potassium during the fruit expansion stage was most effective in preventing fruit shrinkage and increasing pectin content, which were 60.65% lower and 95.4% higher than the control, respectively. Spraying potassium at the expanding and early mature stages helped to maintain the integrity of cuticle and the density and tidy arrangement of the epidermal cells, and reduce the number of cavities in pulp. The effect was the best when spray was made the in expanding stage. There were significant correlations between fruit shrinkage rate and pectin content as well as its catabolic enzyme activity, with a correlation coefficient of 0.997 6 and 0.994 6, respectively. Spraying potassium had little effect on cellulose content and there was no significant correlation between shrinkage rate and cellulose content as well as cellulase activity.【Conclusion】Spraying K_2SO_4 on the leaves during fruit expansion stage is the best way to reduce the occurrence of shrinkage in 'Short branch' winter jujube during maturation. The treatment might effectively promote photosynthesis, increase the protein content, promote the sugar translocation, and thus reduce fruit shrinkage incidence. Based on our study, spraying 0.05 g · mL^{-1} K_2SO_4 every 7 days from July 20 for 3 times can significantly increase the pectin content, enhance the fruit hardness and improve the fiber structure of the fruit, and thus is an effective measure to control fruit shrinkage in 'Short branch' winter jujube.

Key words: 'Short branch' winter jujube; Shrinkage; K_2SO_4; Protopectin; Microstructure

‘短枝’冬枣又称沾化二代冬枣，是山东省滨州市沾化区从一般冬枣自然变异中选育出来的优良冬枣新品种[1]。2010年通过山东省优良品种鉴定。‘短枝’冬枣节间短，果型优美、皮薄肉脆，平均单果质量21.8 g，成熟期含可溶性固形物40%~42%，鲜食品质极好[2]。近几年，山东省、河北省、山西省、陕西省等多处‘短枝’冬枣栽培区出现成熟期果实皱缩、萎蔫现象，有的还伴有叶片提早变黄脱落。一旦发病，果实即失去商品价值。2019年，山东省滨州市沾化区和无棣县的发病率达41%。因未出现果肉褐变和变苦等传统冬枣缩果病的症状[3]，所以‘短枝’冬枣果实皱缩与传统缩果病不是同一种病害。对2016—2018年每年10月上旬采集的皱缩果进行病原菌分离培养，均未培养出病原菌，可见‘短枝’冬枣果实皱缩不是由致病菌引起的病害。

钾元素与果树代谢过程有密切关系，并为多种酶的活化剂，参与糖和淀粉的合成、运输和转化[4-6]。钾能增强原生质体的亲水性和细胞渗透压，能提高树体内糖的储备量和枝干、果实表皮纤维素含量[7]。钾能影响柑橘对钙、镁的吸收[8]，增加柑橘果

皮厚度[9]。施钾可显著提高'阿部白'桃叶片和新梢的K、N、P质量分数、可溶性总糖、还原性糖和淀粉的质量分数；同时果实中的K、N、P质量分数也显著提高[10]。施钾可显著提高'骏枣'果实产量、维生素C含量、可溶性蛋白质含量、糖酸比、可溶性糖含量、蔗糖含量、果糖含量和叶片净光合速率、气孔导度、蒸腾速率、叶绿素含量，且具有钾肥浓度影响效应[11]。我国北方土壤的供钾能力差且自西向东有明显降低的趋势[12]。通过检测发现皱缩果和非皱缩果成熟期的钾含量、果胶及酶、纤维素及酶含量存在差异，因此钾可能与'短枝'冬枣皱缩有一定关系，但二者关联度尚不明确。研究钾对果实皱缩，果胶、纤维素含量及其分解酶活性，果实硬度和显微结构的影响，对揭示'短枝'冬枣果实皱缩发生机制及提出综合防治措施具有重要的指导意义。

1　材料和方法

1.1　试验地概况与试验材料

试验于2018年6—10月和2019年6—10月分别在山东省滨州市沾化区古城镇油坊村冬枣示范园和滨州市无棣县水湾镇现康种植专业合作社冬枣示范园内进行。两园均地处暖湿带半湿润季风性区，气候温和，四季分明，光照充足，雨热同季；区域极端最高气温为41.5 ℃，极端最低气温-23.4 ℃；年平均气温12.5 ℃；年均降水量566.8 mm，主要集中在7月下旬至9月上旬。本试验选取长势一致，树势中等，栽培管理措施相同，由普通冬枣改接5 a(年)以上的盛果期'短枝'冬枣为试验样树。

1.2　试验设计

每个示范园内都采用单因素完全随机实验设计，设置对照、幼果期处理、膨大期处理和白熟期处理，每个处理3次重复，每个重复设置3株树，共36株树。幼果期处理从6月20日开始、膨大期处理从7月20日开始、白熟期处理从8月22日开始。每个时期7 d喷1次0.05 g·mL^{-1}的K_2SO_4溶液，共喷施3次，清水处理作对照。均在傍晚(18:00)喷至叶片滴水为止。其中2次喷药时遇降雨，采用覆盖塑料薄膜的方式进行防风避雨。

于每年10月9日上午7:00—9:00采样。采样部位确定方法：垂直位置选在枣树主干分枝点与顶端的中点上下各15 cm范围内；水平位置为离树中心干30~60 cm处的东、南、西、北4个方位各确定一个采样部位。从每个部位随机选取4个大小一致的枣果，每株树共采果16个。采后立即置于冰盒带回实验室用蒸馏水洗净，置于-20 ℃冰箱中保存，待测。每个重复内所采果实去核粉碎混合后测果胶、纤维素含量及果胶酶、纤维素酶活性。成熟期(10月上旬)调查所选36株树的挂果数、皱缩果数，计算皱缩果率。

1.3　指标测定

果胶含量采用咔唑比色法测定[13]，纤维素含量采取蒽酮法测定[14]，果胶酶及纤维素酶活性参考Brummell等[15]的方法测定。石蜡切片的制作参照贺冰等[16]的方法，OLYMPUS光学显微镜下观察、成像、测量角质层厚度和表皮厚度。果皮硬度和果肉硬度均采用物性分析仪测定，每个果选取赤道部位3个位置进行测定。

1.4　数据分析

采用Duncan's新复极差法(SSR)对结果进行差异显著性检验，利用一元线性回归分析各指标同裂果率间的线性关系。利用SPSS 20.0软件进行统计分析，用Excel制图。

2　结果与分析

2.1　皱缩与非皱缩果果实钾含量、果胶含量、纤维素含量、果胶酶活性和纤维素酶活性比较

经检测分析(表1)，皱缩果果实钾含量、果胶含量、纤维素含量、果胶酶活性和纤维素酶活性均比非

表1　'短枝'冬枣皱缩果与非皱缩果的钾、果胶、纤维素含量及果胶酶、纤维素酶活性

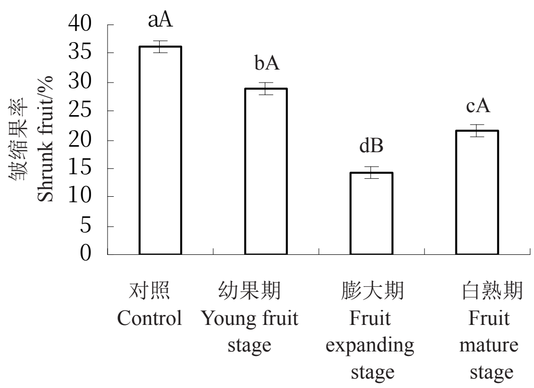
Table 1　Contents of potassium, pectin and cellulose and activities of pectinase and cellulase in shrunken and normal fruit of 'Short branch' winter jujube

果实类型 Fruit type	w(K)/(g·kg^{-1})	w(果胶) Pectin content/(g·kg^{-1})	w(纤维素) Cellulose content/(g·kg^{-1})	果胶酶活性 Pectinase activity/(μg·g^{-1}·min^{-1})	纤维素酶活性 Cellulase activity/(μg·g^{-1}·min^{-1})
皱缩果 Shrunk fruit	2.74±0.24	13.21±0.65	4.82±0.24	79 221±918	16 774±635
非皱缩果 Normal fruit	4.65±0.17	23.75±0.38	6.07±0.24	36 520±774	14 917±706

皱缩果实的含量低。结合皱缩枣果的症状和病原菌培养情况，初步将‘短枝’冬枣皱缩认定为是一种因缺钾引起的生理病害。

2.2 不同时期喷钾处理对‘短枝’冬枣皱缩率的影响

‘短枝’冬枣坐果后各时期叶面喷施0.05 g·mL^{-1} K_2SO_4均能显著降低皱缩果率。从图1中可看出，降低效果依次为膨大期喷钾>白熟期喷钾>幼果期喷钾>对照。幼果期、膨大期、白熟期喷钾后冬枣皱缩率分别比对照降低了20.18%、60.65%和40.09%，幼果期喷钾效果显著，膨大期喷钾后效果极显著。表明叶面喷施K_2SO_4有助于‘短枝’冬枣皱缩率的下降，并且在膨大期喷施效果最好。



不同小写字母表示差异显著($p < 0.05$)，不同大写字母表示差异极显著($p < 0.01$)。下同。

Different small and capital letters indicate significant difference at $p < 0.05$ and $p < 0.01$, respectively. The same below.

图1 不同时期喷钾处理对‘短枝’冬枣皱缩率的影响

Fig. 1 Effect of K treatment on fruit shrinking rate in ‘Short branch’ winter jujube

2.3 不同时期喷钾处理对‘短枝’冬枣果实果胶酶活性和果胶含量的影响

叶面喷施K_2SO_4能增加‘短枝’冬枣果实的果胶含量(图2)。坐果后到白熟期喷钾的时间越晚果胶含量越高。与对照相比，幼果期、膨大期和白熟期喷钾，对果实果胶含量分别增加了34.1%、95.4%和60.3%。幼果期喷钾效果不显著，白熟期喷钾效果显著，膨大期喷钾效果极显著。

叶面喷施K_2SO_4能显著降低‘短枝’冬枣果胶酶活性(图3)。幼果期、膨大期和白熟期处理的‘短枝’冬枣中果胶酶活性分别降低了21.6%、76.5%和53.1%。幼果期喷钾后果胶酶活性与对照差异显著，膨大期和白熟期差异极显著，与果胶含量的变化

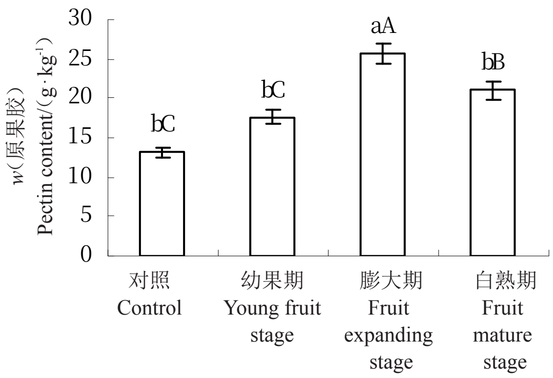


图2 不同时期喷钾处理对‘短枝’冬枣果胶含量的影响

Fig. 2 Effect of K treatment on pectin content in ‘Short branch’ winter jujube

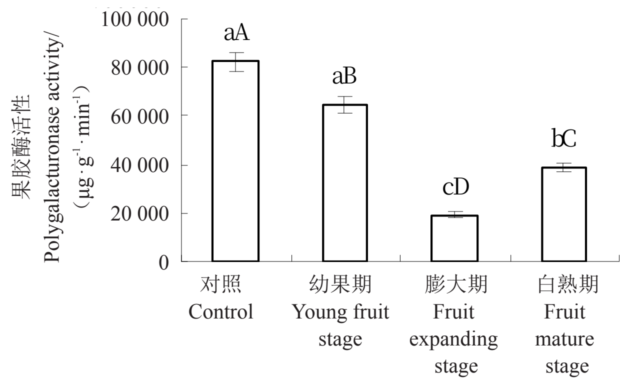


图3 不同时期喷钾处理对‘短枝’冬枣果胶酶含量的影响

Fig. 3 Effect of K treatment on polygalacturonase activity in ‘Short branch’ winter jujube

趋势相反。上述结果表明从幼果期到白熟期喷钾的时间越晚越能有效抑制果胶酶活性，减缓果胶的降解速度，最终增加成熟期冬枣的果胶含量。

2.4 不同时期喷钾处理对‘短枝’冬枣果实纤维素和纤维素酶活性含量的影响

叶面喷施K_2SO_4也能增加‘短枝’冬枣的果实纤维素含量(图4)。幼果期、膨大期和白熟期喷钾，果实中纤维素含量分别增加了43.1%、21.2%和10.7%，仅在幼果期喷钾达到显著水平，膨大期和白熟期喷钾对果实纤维素含量影响均不显著。

叶面喷施K_2SO_4能显著降低‘短枝’冬枣果实纤维素酶活性(图5)。与对照相比，幼果期、膨大期和白熟期喷钾，果实中纤维素酶活性下降了87.3%、88.4%和64.0%，各时期喷钾后果实中的纤维酶活性显著降低，且幼果期喷钾后果实中的纤维酶活性极显著降低，表明喷钾处理显著抑制了纤维素酶活性。

2.5 枣皱缩果率与各指标的相关性

‘短枝’冬枣果实皱缩率与果胶含量、果胶酶活

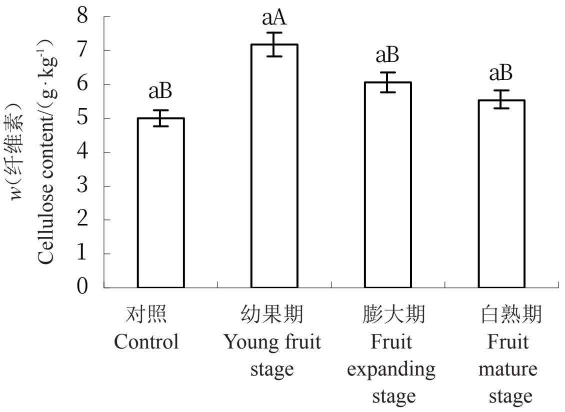


图4　不同时期喷钾处理对‘短枝’冬枣纤维素含量的影响

Fig. 4　Effect of K treatment on cellulose content in ‘Short branch’ winter jujube

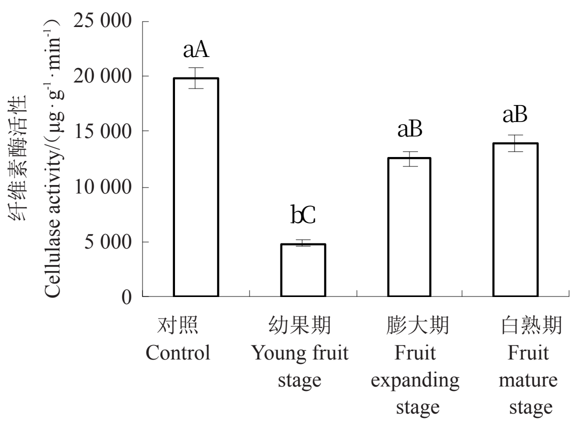


图5　不同时期喷钾处理对‘短枝’冬枣纤维素酶活性的影响

Fig. 5　Effect of K treatment on cellulase activity in ‘Short branch’ winter jujube

性2项指标存在显著相关性（表2）。果胶酶活性与皱缩率呈极显著正相关，果胶与皱缩率呈极显著负相关。‘短枝’冬枣喷施K_2SO_4后冬枣果实内维持细胞壁稳定的果胶含量含量提升，皱缩率降低。其中又以膨大期喷施K_2SO_4防止果实皱缩效果最明显。

表2　‘短枝’冬枣皱缩率与果胶、纤维素含量及其降解酶活性的相关性

Table 2　Correlations between the shrinkage rate of ‘Short branch’ winter jujube and the contents of pectin and cellulose and the activities of their degrading enzyme

指标 Index	方程 Equation	系数 Coefficient
果胶含量 Protopectin content	y=−0.560 9 x+33.469	0.997 6**
果胶酶活性 Polygalacturonase activity	y=2 942 x−23 032	0.994 6**
纤维素含量 Cellulose content	y=−0.021 7 x+6.492 7	0.049 2
纤维素酶活性 Cellulose activity	y=178.69 x+8 281.4	0.074 1

注：**表示极显著相关 $p < 0.01$。

Note: **indicate significant correlation at $p < 0.01$.

‘短枝’冬枣果实皱缩率与纤维素含量、纤维素酶活性的相关性不显著。

2.6　不同时期喷钾处理对‘短枝’冬枣果实硬度的影响

不同时期喷钾果皮硬度为6.54~7.94 kg·cm^{-2}，果肉硬度2.27~2.81 kg·cm^{-2}。不喷钾果皮硬度为5.92 kg·cm^{-2}，果肉硬度为2.14 kg·cm^{-2}。膨大期喷钾果皮和果肉硬度最高硬度为(7.94±0.57)kg·cm^{-2}和(2.81±0.43)kg·cm^{-2}。喷钾可使果皮、果肉硬度显著大于对照，其中又以膨大期喷钾效果最为明显。膨大期和白熟期喷钾可使果肉硬度显著大于其他处理，幼果期喷钾的枣果果肉硬度与对照无明显差异。膨大期与白熟期喷钾对果皮硬度的影响差异显著，但对果肉硬度的影响差异不大（表3）。由此可见，膨大期喷钾对提高果实硬度的效果最明显。

表3　不同时期喷钾处理对‘短枝’冬枣果实硬度的影响

Table 3　Effect of potassium treatments on fruit hardness in ‘Short branch’ winter jujube　（kg·cm^{-2}）

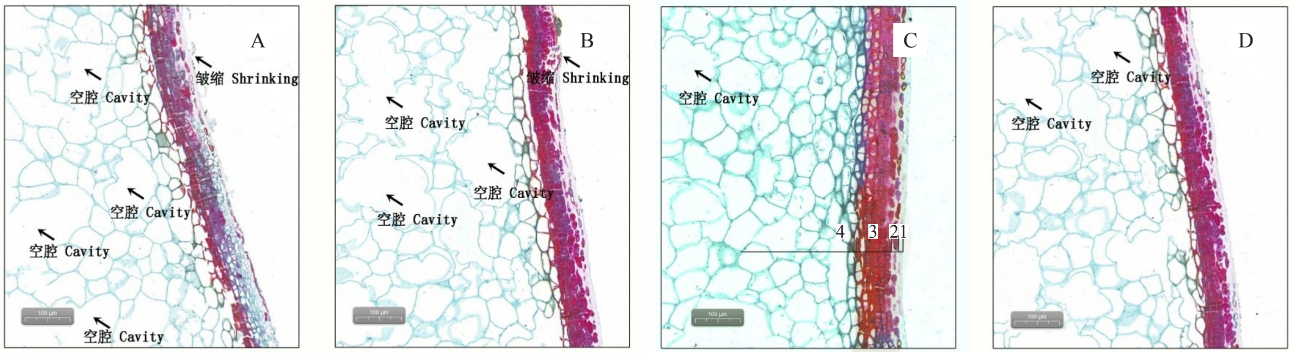
部位 Position	喷钾时期 Potassium spraying time			
	对照 Control	幼果期 Young fruit stage	膨大期 Expending stage	白熟期 Maturation stage
果皮 Peel	5.92±0.97 c	6.54±1.02 b	7.94±0.57 a	7.17±0.62 b
果肉 Flesh	2.14±0.54 b	2.27±0.47 b	2.81±0.43 a	2.61±0.35 a

2.7　不同时期喷钾处理对‘短枝’冬枣果实显微结构的影响

喷钾后‘短枝’冬枣果实显微结构形态特征如图6所示。空白对照角质层发生龟裂最严重。K_2SO_4可使枣果实角质层完整，不发生龟裂，以膨大期喷施效果最明显；对照表皮细胞较大，排列疏松无序，空腔多，分布不均匀。钾处理后果肉空腔数有所减少；各处理枣果实表皮细胞尺寸由小到大和排列紧实规整程度为膨大期>白熟期>幼果期>对照。

3　讨　论

钾能增强原生质体的亲水性，能有效提高果实果胶含量和抗皱缩能力。果胶是存在于高等植物初生细胞壁和中胶层中的一种酸性多糖物质，其代谢与果实质地变化密切相关。近年来，各种细胞壁果胶物质如β-半乳糖苷酶、β-甘露糖苷酶和α-呋喃阿



A. 对照；B. 幼果期；C. 膨大期；D. 白熟期。1. 角质层；2. 表皮层细胞；3. 亚表皮层细胞；4. 果肉细胞。

A. Control; B. Young fruit stage; C. Fruit expanding stage; D. Fruit maturation stage. 1. Cuticle; 2. Epidermal cell; 3. Subepidermal cell; 4. Fresh cell.

图 6 不同时期喷钾处理对'短枝'冬枣果实显微结构的影响

Fig. 6 Effect of K treatment on the microstructure of 'Short branch' winter jujube fruit

拉伯糖苷酶等在果实细胞壁成分降解和质地软化中的作用得到越来越多的研究[17-19]。细胞壁不同组分往往以糖苷键相连接而牢固地结合在一起，维持了细胞壁的机械强度。细胞壁糖苷酶能够水解这些糖苷键，释放出中性糖，促进细胞壁物质的解聚和溶解。有研究表明，随着果实的成熟，细胞壁中的WSP（water soluble pectin）发生降解[20]。

本试验中，不同时期叶面喷K_2SO_4不同程度地降低了'短枝'冬枣成熟期的果实皱缩率，降低幅度依次为膨大期>白熟期>幼果期，膨大期处理效果最显著降低了60.65%。喷钾后果实的钾、果胶、纤维素含量显著升高，果胶酶活性下降，这一结果验证了试验初期的初步认定结果。冬枣膨大期甲口刚刚愈合，枣果生长、根系恢复以及枝叶生长均需要较多的营养供给，是冬枣营养缺乏、树势较弱、最易受病虫害侵袭的时期。此时叶面喷K_2SO_4有效促进光合作用，提高果品蛋白质含量，促进果实糖分转化，进而降低果实皱缩率。这与前人在柑橘[8-9]和'骏枣'[11]上的研究结果是一致的。还有研究表明，宁夏'同心圆枣'树萌芽后应注重氮肥的施用，随着生育期的推移，应逐渐增加磷、钾肥的施用[21]与本研究得出的'短枝'冬枣对钾的需求规律一致。果胶含量与'短枝'冬枣皱缩率显著正相关，相关系数达0.997 6。'短枝'冬枣成熟期发生果实皱缩的原因可能是果胶含量低，进而细胞结构受损果肉空腔变大变多最终导致果实质地软化[22]。喷钾后随着细胞壁结构的改善，果实硬度相应增强，皱缩率也显著下降。这与苹果发育过程中果胶含量的趋势是一致的[23]。

4 结 论

不同时期叶面喷施K_2SO_4均能降低'短枝'冬枣成熟期的皱缩率，且膨大期喷施效果最好。膨大期和白熟期喷钾显著抑制了果胶酶活性，降低了果胶的降解程度，增加了果胶含量。果胶含量与果实皱缩率显著相关。虽然喷钾也抑制了果实纤维素酶活性，对纤维素含量有一定影响，但纤维素含量与果实皱缩率关系不密切。膨大期和白熟期喷钾对提高果实硬度的效果最明显，也可有效改善果实显微结构。因此，7月20日至8月22日每隔7 d喷1次，共喷施3次0.05 $g\cdot mL^{-1}$ K_2SO_4可显著增加果实果胶含量，增强果实硬度，改善果实纤维结构，是防治'短枝'冬枣成熟期果实皱缩的有效措施。

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