1 2	Study on seedling cultivation using buds of potato (Solanum tuberosum L.)
2 3 4	Chaonan Wang <sup>1,2</sup> , Chong Du <sup>1,2</sup> , Zhongmin Yang <sup>1,2</sup> , Huilin Wang <sup>2</sup> , Leijuan Shang <sup>2</sup> , Lili Liu <sup>2</sup> , Zhiyi Yang <sup>2</sup> , Shuyao Song <sup>3</sup> , Sikandar Amanullah <sup>4</sup>
5	Dru , Zhiyi Tung , Shujuo Song , Shuhun Timunuhun
6	<sup>1</sup> Postdoctoral Station of Horticulture, Xinjiang Agricultural University, Urumuqi, 830052, PR
7	China
8	<sup>2</sup> College of Horticulture, Xinjiang Agricultural University, Urumuqi, 830052, PR China
9	<sup>3</sup> College of Horticulture, Jilin Agricultural University, Changchun, PR China
10	<sup>4</sup> College of Horticulture and Landscape Architecture, Northeast Agricultural University, Harbin,
11	150030, PR China
12	
13	Corresponding Author:
14	Chong Du
15	<sup>1</sup> Postdoctoral Station of Horticulture, Xinjiang Agricultural University, Urumqi, Xinjiang
16	830052, PR China
17	<sup>2</sup> College of Horticulture, Xinjiang Agricultural University, Urumqi, Xinjiang 830052, PR China
18 19	Email address: godv2018@163.com
20	Zhongmin Yang
21	<sup>1</sup> Postdoctoral Station of Horticulture, Xinjiang Agricultural University, Urumqi, Xinjiang
22	830052, PR China
23	<sup>2</sup> College of Horticulture, Xinjiang Agricultural University, Urumqi, Xinjiang 830052, PR China
24	Email address: <u>yangzhongmin161220@126.com</u>
25	
26	Abstract
27	Background: The world is facing numerous challenges in food production due to unstable
28	climate change. There is a strong need to develop the various techniques of crop production,
29	which could greatly help fulfil the demand for better crop production within a short time period.
30	Methods: In this study, a bud planting method was adopted to cultivate potato seedlings. For this
31	purpose, we assessed different types of treatments for producing quality buds and seedlings of
32	potatoes. Total 4 disease-free potatoes (Fujin, Youjin, Zhongshu 4, and Feiwuruita) were
33	selected. Potato buds with three different lengths (3cm, 5cm, and 7cm) were considered T1, T2,
34	T3 treatments, and terminal buds, middle buds, and tail buds were used as T4, T5, and T6
35	treatments, respectively. The experiment was performed in nutrient pots using a randomised
36	complete block design (RCBD) and replicated thrice compared with the natural control (CK)
37	treatment. The cultivation was done by adopting the common horticultural practices of weeding
38	and hoeing as needed for growing conditions. The photosynthetic indexes, physiological indexes,
39	growth indexes and quality of potato seedlings, potato bud quality were measured at two weeks

40 intervals, and yield indexes were measured when the final crop was harvested 14 weeks after41 planting.

42 **Results and Conclusions:** The obtained results depicted that seedling cultivation of potato bud

43 with different lengths increased the reproduction coefficient as well as reduced the number of

44 seed potatoes used for cultivation. All the morphological, physiological and yield indexes were

45 also improved in a positive trends. The 7cm potato bud length was proven as the best treatment

among the different treatments used for raising seedling. Different parts of potato bud alsoshowed the formation of the best quality bud at the top section of the potato. In crux, we

47 showed the formation of the best quality bud at the top section of the potato. In clux, we

48 hypothesised that our proven bud planting technique should be adopted commercially, which

- 49 could help in good crop production with maximum yield.
- 50

51 Key words: Potato; Seedling cultivation; Production; Potato bud; Yield

52

# 53 Introduction

54 Potato (Solanum tuberosum L.) isa well-known tuber crop around the world, and approximately 55 1.3 billion people consume this crop as a staple food (Stokstad, 2019). Potatoes are native to the Andes mountains of South America, with an altitude of 2000-4000 M. The environmental 56 57 characteristics are short daytime, high light intensity, low temperature and high relative humidity (Harris, 2012). After hundreds of years of domestication, this crop was primarily introduced into 58 59 Europe in the 1970s. It is being cultivated in several countries and regions between 65°N and 50°S of the world (*Camire et al., 2009*). This crop is also used as an important industrial raw 60 material for the production of starch, ethanol and animal feed (Birch et al., 2012). 61

A potato tuber is a modified stolon which is a rich source of nutrition intake due to stored starch, protein, vitamins and minerals (*Aien et al., 2011; Hancock et al., 2014*). This tuber is an expanded stem with terminal buds and lateral buds, and these buds enter into a dormant state after separating from the mother part. Tuber formation begins with the inhibition of longitudinal growth at the end of stolon, and then the terminal bud begins to expand due to cell division and cell expansion (*Cutter, 1978*). The formation of potato tuber is generally depended upon favorable climatic conditions.

69 A number of different planting techniques have been introduced to get suitable potato 70 production. Among them, potato bud planting technology achieved the purpose of restoring 71 seeds with virus-free and strong disease resistance characters at an early stage. When the late 72 blight occurs in rainy season of July/August, then potato crop is being to be harvested or close to its maturity level (Luo and Jin, 1960). The breaking off potato buds dormancy, raising of 73 seedlings in the seedbeds, and transplanting them to proper field for rapid seed propagation could 74 75 save up to 39.3% of seed potatoes amount (*Wu et al., 2009; Li, 2014*). When the seedling reach 76 at 5 cm height, it bear a large number of fibrous roots in the soil, then it should be detached from 77 the seed potato for transplanting, which could increase the reproduction coefficient of virus-free 78 seed potato (Xiong et al., 2010).

79 The cultivation of potato bud seedlings is mainly linked with the potato planting method of

- 80 seedling transplanting, which is primarily based on the cultivation of potato bud as seed. Bud
- 81 planting technology has the characteristics of multiple reproduction, fast reproduction, and
- 82 guaranteed crop cultivation, and this method been used in traditional Chinese medicine crops (Yu
- 83 *et al., 1999; Wen et al., 2000*) and vegetable crops (*Li, 1962*). The potato bud planting method
- 84 was introduced in 1960s and has been tested in different provinces (Shanxi, Zhejiang, Jilin,
- 85 Heilongjiang, Qinghai and other areas) of China (Li, 1959), respectively. The planted potatoes'
- buds are accelerated in semi-humid soil under a suitable temperature of 20-25°C. The length of
  speedy germination can reach 3.5-10.5cm growth in 8 days, and the survival rate of transplanting
- is relatively high (*Liu*, 1962). When seedlings reached at 6-10cm height and 5-7 leaves numbers,
  then seedlings were cut off from the base and planted in time. The yield was higher than
- 90 traditional direct seeding with buds (*Zhang and Tian, 2012*).

91 It has been shown that when the bud length of seed potato reaches 2.5cm, crop production with the method of 2.5cm length bud could increase up to 89.4% compared to the conventional 92 method of direct potato seeding (Lv and Jiang, 1992). The cultivation of potato seedlings was in 93 94 favor of reducing the occurrence of potato late blight disease and also reduced the probability of 95 plants degradation (Chen, 2014; Chen and Jiang, 2015). A study showed that the much older seed tuber buds produce a higher yield. The bud planting method saves the number of seed 96 potatoes as compared with the direct planting method (Liu et al., 1990). The traditional way of 97 98 natural bud seeding yield 200-300 kg/mu of potatoes, but the cultivation through the budseedling planting method only need 40-50 kg/mu, which could save nearly five times seed 99 100 potatoes (Li, 1982).

101 The raising of the vegetable seedlings is primarily linked with vegetable cultivation (Wen and Li, 2001). Seedling cultivation is one of the fundamental measures to check and improve the 102 103 emergence rate and cultivate strong seedlings during vegetable production. It has numerous 104 advantages of enhancing the precocity of vegetables, increasing yield, and increasing the 105 economic income for farmers. At present, potato's protected culpotatoeson mainly ddependent upon the direct seeding of tubers, which has the crucial disadvantages of large seed consumption 106 107 and low reproduction coefficient (Fig. 1A). However, using the potato buds for seedling 108 cultivation can effectively reduce its seed consumption, increase the reproduction coefficient and 109 increase the overall efficiency of the crop. Very little is reported on potato seedling cultivation 110 through buds usage. So, the undertaken experiment laid the adaptability and feasibility of potato 111 seedling cultivation using the different buds length in protected nutrient pot. The authors found 112 that potato bud-seedling cultivation is conducive to early maturity and yield improvement.

113

#### 114 Materials & Methods

115

# 116 Source of material & cultivation

- 117 Total four virus-free potatoes (Fujin, Youjin, Zhongshu 4, and Feiwuruita) were used as
- 118 experimental varieties which were collected from Jilin Academy of Agricultural Sciences, China.

119 These materials were healthy seed potatoes having excellent phenotypes of diameter, height,

120 shape, and good bud development. The cultivation of these experimental materials was grown in

a greenhouse of a practical experiment farm of Jilin Agricultural University, China. The sandy

loam soil was arranged, and farm manure was mixed in it. The mixed soil was filled in nutrient

123 up to 30cm of depth, pot The physical and chemical properties of soil were ranged comprising of

total nitrogen 1.41 g/kg, total phosphorus 0.34 g/kg, total potassium 8.9 g/kg and organic matter
40.6 g/kg.

125 126

# 127 Experiment design and measurements

The seed potatoes were selected on the basis of their significant morphological characteristics and without any visual abnormality. Three independent trials were carried out in all the cases.

130 (1) Raising of seedlings and cultivation of different potato bud lengths: The length of potato 131 bud was divided into three treatments of 3cm±0.5cm, 5cm±0.5cm, and 7cm±0.5cm, respectively. The virus-free seed potato of the same size, and strong dormancy period were used to promote 132 133 the germination under dark light period. When the bud length reaches at 3cm±0.5cm (seed potato 134 treatment for 8-9d, T1), 5cm±0.5cm (seed potato treatment for 10-11d, T2) and 7cm±0.5cm 135 (seed potato treatment for 12-13d, T3) (Fig. 1B), then potato terminal buds were selected and 136 bud breaking treatment was done. The buds were broken from basal ends, sown into nutrient 137 bowl having equal size of 10cm×10cm. Seedlings were successfully raised in the greenhouse on 138 March 25, and 25 days old seedlings were then planted in the greenhouse on April 20.

(2) Raising of seedlings and cultivation of different parts of potato buds: Three treatments of
terminal bud (T4), middle bud (middle part of seed potato, T5) and tail bud (basal bud, T6) were
set at different sections of potato bud. When the length of the potato bud was 5cm±0.5cm (seed
potato treatment for 10-11d) (Fig. 1C), then bud breaking treatment was carried out from the
basal part of the potato bud, and the other methods were the same as described in the above part
1.

145 (3) The cutting of seed potato seedling and cultivation in a nutrient bowl (CK) (Fig. 1A): The 146 seed potato was taken out from the storage pit and placed in an incubator light under 20°C 147 temperature and 60% humidity. When the bud length reached 1-2cm, then the bud was cultured under indoor scattered light for 2~3 days. The seed potato was cut into bud blocks having 20g 148 149 weight, sown into the same nutrition bowl of 10cm×10cm, and one plant per bowl was 150 successfully developed. The seedlings were raised in greenhouse on 17 March, and when the 151 seedling age was 25 days old, then transferred into the greenhouse on 2 March, all seedlings grown together. 152

Total three experimental trials were collectively cultivated in raised beds of 1.2m width, 5m length, 10cm height and protective transparent film coverage was applied. For double row cultivation, the row spacing of plants on the border was 25cm×40cm. The experiment was repeated 3 times and arranged in randomised complete block designs (RCBD) manners. All the agronomic practices, e.g., watering, fertilisation, temperature, and humidity, were maintained according to the needs and production routine. 159

#### 160 Sampling and measurements

161

#### 162 Measurement of photosynthesis rate and chlorophyll contents

163 The photosynthesis rate of potato seedlings was determined at suitable time of 9:30 am to 11:30

am. Ten leaves samples (each sample was third mature leaflet from top to the bottom of plant)

165 were carefully selected for photosynthetically active radiation (PAR). Then, photosynthetic rate

166 (Pn), transpiration rate (Tr), and stomatal Conductance (Gs) was determined by using a LI-

167 6400XT photosynthetic apparatus (LI-COR, Inc., Lincoln, NE, USA). The illumination intensity

168 was 1,600  $\mu$ mol photons m<sup>2</sup>/s, and gas flow rate was 500 mmol/s, concentration of cuvette CO<sub>2</sub>

169 was set at 400 mmol  $CO_2/mol$  air, and the chamber temperature was 28°C. Then, the mixture was

170 centrifuged with 85% (v/v) acetone solution for chlorophyll extraction, and spectrophotometric

171 (HALO DB-20, Calamb, UK) measurements were obtained at 663 and 645 nm (*Paul and* 

172 *Driscoll, 1997*). The leaves of potato were sampled for determination of total chlorophyll

- 173 contents in plants.
- 174

## 175 Measurement of agronomic parameters

176 The diameter of stem was measured from 5 replicates of standing field plant by using an

177 electronic vernier calliper (instrument precision, 0.01 mm) as mentioned earlier (*Chang et al.*,

178 2016). The length and height of the potato stem were measured using the measuring ruler having

a 1 mm scale. A total of 5 plants were selected from each treatment, readings were taken 3 times,

and the average value was calculated for determining the number of compound leaves per plant.

181 The area of the compound leaf was measured at the seedling stage and tuber expansion stage. For

this purpose, 5 plants were randomly selected and measured from each treatment for 3 times by

using a handheld laser leaf area meter (Ci-203, CID, Inc., Vancouver, WA, USA).

Potato plants were harvested for the measurements of agronomic parameters. The aboveground biomass was earthen up and measured after plants were oven-heated at 70°C temperature.

186 The dry matter of plant organs (leaves, stems, roots, stolons and tubers) were weighted after

187 detaching them from plants. The ratio of organs dry matter = the organ (leaf, stem, root, stolon,

detaching them from plants. The facto of organs dry matter – the organ (lear, stelli, foot, store) 100 – the plant (lear, stelli, foot, store)

tuber) weight / the above ground biomass×100%; the ratio of root shoot = the root dry weight /
the shoot dry weight×100%; the sound seedling index = (stem diameter / plant height + root dry

190 weight / shoot dry weight)  $\times$  (root dry weight + shoot dry weight).

191 Root activity was analysed by the triphenyl tetrazolium chloride (TTC) method (*Wang et al.*,

192 2006). TTC is a chemical that is reduced by dehydrogenases, and mainly succinate

193 dehydrogenase when added to a tissue. The dehydrogenase activity is regarded as an index of the

root activity. In brief, 0.5 g of fresh root was immersed in 10 ml of an equally mixed solution of

195 0.4% TTC and phosphate buffer, and kept in the dark at  $37^{\circ}$ C for 2 h. Afterward, 2 ml of H<sub>2</sub>SO<sub>4</sub>

196 (1 mol/L) was added to stop the reaction with the root. The root was dried with filter paper and

197 then extracted with ethyl acetate. The red extractant was transferred into the volumetric flask to

reach 10 ml by adding the ethyl acetate. The absorbance of extract at 485 nm was recorded.

Finally, root activity was expressed as TTC reduction intensity. Root activity = amount of TTC reduction  $(\mu g)$  / fresh root weight  $(g) \times time$  (h).

201

## 202 Determination of potato yield and quality

After the harvesting, the total tubers at each sub-plot were weighed for yield assessment using an electronic scale, and the number of tubers per plant was counted. Relative yield = total yield of each treatment / yield of CK. The starch content of the plant sample was assayed as reported earlier (*Grechi et al., 2007*) method. After removing the plant residue, tissue starch was extracted by 80% (v/v) ethanol. After adding 3% HCl to the residue, the spectra were determined by photometric determination at 490 nm by the phenol-sulfuric acid method. A glucose calibration curve was established to calculate tissue starch content, expressed in mg/g dry weight.

For determination of reducing sugar, each reducing sugar sample was collected and placed into three test tubes with pistons. The filtrates of centrifuged potato tuber was grinded into liquid (0.2 mL), and distilled water (1.8 mL) were added to the test tubes. The determination method of

reducing sugar was the DNS (*Miller, 1959*) method. The recorded data was substituted into a

standard curve to calculate the reducing sugar content of grinded potato tuber liquid. The crude

215 protein was detected by Coomassie brilliant blue staining (*Gao, et al., 2014*), and vitamin C was 216 determined by molybdenum blue colourimetry (*Li, 2000*).

217

#### 218 Data analysis

219 The recorded data was statistically analyzed by using the statistics program SPSS 20.0 (SPSS

- Inc., Chicago, IL, USA). A one-way analysis of variance (ANOVA) with posthoc tests (Tukey)
  was used to establish differences among treatments. The mean values showing significant
- differences were compared with the Tukey test at a 5% level.

#### 224 Results

# 225

223

# 226 Effects of seedling raising on physiological indexes and growth indexes of potato at seedling227 stage

The chlorophyll content and net photosynthetic rate of developed seedlings with different tuber bud lengths showed the highest values at T1, followed by T2, and T3 showed the lowest values,

but they were significantly different from those treated with CK, as shown in Table 1. The potato

seedlings were obtained by using different parts of potato buds to cultivate seedlings in nutrient

- bowls, and the results of physiological indicators of each seedling stage were analysed and
- shown in Table 2. The chlorophyll content and net photosynthetic rate were responsive to the
- following treatments T4>T5>T6>CK, and T4 and T5 treatments were significantly different
- from T6 and CK, respectively, but no significant difference was observed between T6 and CK.
- The net photosynthetic rate determination was not significant for nursery treatment of T5, T6 and
- 237 CK. The transpiration rate and stomatal conductance decreased in turn in the four treatments of
- the same variety, and the tested potato varieties showed consistency.

- 239
- 240 Effects of seedling raising on growth indexes and quality of potato plants at seedling stage

There were significant differences in plant height, stem diameter, and number of compound
leaves of potato at seedling stage among the treatments with different length of potato bud (Table

- 3). The seedling cultivation with 7cm potato bud was noticed as the longest seedling ascompared to 5cm seedling treatment and 3cm seedling treatment, and the difference was
- significant; however, overall three treatments results were much higher than CK treatment.
- Regarding the plant height, stem diameter and number of leaves, T1 treatment of 'Fujin' 246 247 variety increased by 49.63%, 60.94% and 58.81% as compared with CK, respectively. The T1 248 treatment of 'Youjin' increased by 21.80%, 57.83% and 43.61% as compared to CK; and T1 treatment of 'Zhongshu 4' was 49.12%, 51.09% and 40.93% higher than that of CK; then T1 249 250 treatment of 'Feiwurita' increased by 37.38%, 44.7% and 35.69% with CK. The quality 251 difference of potato seedlings at seedling stage under the treatment of different potato bud 252 lengths was as followed; the above-ground dry weight, underground dry weight, root-to-shoot 253 ratio, root vigor, and strong seedling index of different potato bud length were T1>T2>T3>CK. 254 The potato bud seedlings of three lengths were significantly different from CK treatment (Table 255 4).
- 256 Further, different parts of potato buds were used for seedling cultivation in a nutritional bowl 257 and research results at seedling stage are shown in Table 5. The plant height, stem diameter and a number of compound leaves of each treatment in the same variety showed T4>T5>T6>CK, and 258 259 the potato terminal bud treatment was higher with significant differences as compared to the CK 260 levels. All the tested varieties showed the same performances, and the indexes of plant height, 261 stem diameter and leaf number of 'Fujin' increased by 32.10%, 43.94% and 35.29%, and "Youjin" increased by 11.01%, 48.44% and 22.95%, respectively, 'Zhongshu 4' increased by 262 263 31.40%, 35.62% and 20.37%, 'Feiwurita' increased by 20.00%, 27.85%, and 17.24%, and all of 264 above increments are very much as compared with CK treatment levels, respectively.
- The effects of potato bud seedling raising at different parts on potato seedling quality are shown in Table 6. It can be seen that the above-ground dry weight of seedling raising treatment was higher than CK level, and four varieties depicted consistent results. The dry weight of the above ground and the dry weight of the underground part of the potato top bud seedling cultivation treatment was significantly different from that of CK. The root activity and seedling index of all tested varieties were T4>T5>T6>CK, and the performance trend of all varieties was the same.
- 272

#### 273 Effect of seedling cultivation on potato plants in greenhouse

By comparing the plant growth of potatoes treated with different lengths of potato buds duringthe tuber expansion stage, it was found that three growth indicators of plant height, stem

the tuber expansion stage, it was found that three growth indicators of plant height, stemthickness, and leaf area in each treatment and within the variety were different, as followed by

- 277 the children in each treatment and within the variety were different, as followed by 277 T1 T2 T2 CK T1 to the theory is  $\frac{1}{100}$   $\frac{1}{100}$   $\frac{1}{100}$   $\frac{1}{100}$   $\frac{1}{100}$
- T1>T2>T3>CK. The treatments were significantly differed and performance trends of four
- experimental varieties were consistent.

279 Among the all treatments, the plant height increased at maximum level of 37.63% in T1 280 treatment of 'Feiwurita' variety and the stem thickness and leaf area were increased the most in T1 treatment of 'Feiwurita', which were 37.32% and 16.84% respectively (Table 7). The 281 282 comparison of plant growth in potato buds at different positions during the expansion period of 283 potato tuber showed from Table 8. Meanwhile, in the potato tuber expansion period, the three growth indicators of plant height, stem thickness, and leaf area were all shown as follows for 284 each variety treatment T4>T5>T6>CK. Among them, the highest plant height was 69.52cm in 285 potato top bud seedling cultivation of variety 'Eugene', the maximum stem thickness was 286 287 15.99mm for 'Zhongshu 4', and the leaf area of 'Zhongshu 4' was the highest as 6810cm<sup>2</sup>. By 288 comparing with the control plant height, the highest increase in stem thickness was the 289 cultivation of 'Fujin' potato top bud cultivation, increasing by 24.31%, 31.28%, and 13.55% 290 (Table 8).

291 During the expansion stage of potato tuber, the dry matter distribution rate of potato tuber in 292 different tissues and organs was the highest, and order from large to small was tuber, stem, leaf, 293 root and stolon. The distribution rate of dry matter of potato bud seedling treatment with 294 different lengths was T1>T2>T3>CK. The distribution rates of tissues and organs of potato 295 tuber, stem and leaf in T1 and T2 of the same variety were basically the same, with little 296 difference, but significantly higher than T3 and CK treatment. In the T1 treatment cultivation of 297 various varieties, the proportion of tubers was about 33%, and the proportion of stems and leaves 298 was about 64%.

299 Among them, the proportion of 'Zhongshu No. 4' tubers was the highest (34.12%), and 300 proportion of stems and leaves was 63.01%. The stem and leaf were in the main proportion in 301 CK, and the highest proportion of 'Youjin' was 80.87%. The dry matter distribution rate of the 302 potato in the tuber expansion stage under the cultivation of different parts of the potato buds was 303 shown as the dry matter distribution rate of the tuber T4>T5>T6>CK. The 4 tested varieties 304 showed the same performance, and the tuber stem of 'Zhongshu 4'. The highest material 305 distribution rate was 34.12%, and the lowest rate was found 16.30% for the direct seeding 306 treatment of 'Youjin' bud block. The stem dry matter distribution rate and leaf dry matter 307 distribution rate of each variety were the highest in CK (Fig. 2).

308 The comparison of physiological indexes of various experimental treatments in potato tuber 309 expansion period showed that under the seedling raising level of potato bud nutrient bowl with 310 different lengths, the chlorophyll content and photosynthetic indexes of potato plants in the same variety were the highest in T1 treatment, as followed by T2>T3 treatment, and the lowest was in 311 312 CK. The maximum photosynthetic rate was 20.9% in 'Feiwurita' T1 treatment µMolCO<sub>2</sub> •m<sup>-2</sup>•s<sup>-</sup> <sup>1</sup>, the highest chlorophyll content was found in the T1 treatment of 'Zhongshu 4', which was 313 314 4.26 mg/g; Compared with potato seedling, the chlorophyll content of four tested varieties 315 "Fujin, Youjin, Zhongshu 4 and Feiwuruita" increased at the utmost level, which were 32.97%, 316 49.57%, 51.06% and 37.93%, and the photosynthetic rate increased by 23.33%, 23.81%, 22.22% 317 and 15.47% respectively (Table 9).

318 The research on physiological indexes of different parts of potato bud during the expansion 319 period of potato tuber showed that there were significant differences in chlorophyll content, 320 photosynthetic rate, and transpiration rate of other parts of potato sprouts during the tuber 321 expansion period (Table 10). The photosynthetic rate and transpiration rate of potato during the 322 expansion stage of potato tuber showed the order of T4> T5> T6> CK, and T4 and T5 were 323 significantly different from T6 and CK, respectively. The performance trends of all varieties 324 were seemed same. Among them, the chlorophyll content of 'Zhongshu 4' with the highest rate of 3.89mg/g, and highest photosynthetic rate of 'Feiwurita' is 20.1µmolCO<sup>2</sup>•m<sup>-2</sup>•s<sup>-1</sup>. The 325 stomatal conductance of different parts of tuber buds during expansion period was not obvious. 326 327 The chlorophyll content of tuber top bud seedling treatment of 'Zhongshu 4'. As compared with 328 CK, the photosynthetic rate increased most obviously, increasing by 37.94% and 16.67%, 329 respectively.

330

#### 331 Effects of seedling cultivation methods on earliness and yield of potato

The planting date of each treatment was the same, and the harvest time and growth days in the shed were different. The earliest maturity among treatments in the same variety was T1, followed by T2, T3 and CK. Compared with the CK, T1 treatment could harvest 13 days in advance, and the difference of harvest days between T1 and T2 treatment is small, only 1-2 days (Table 11)

The yield composition of all tested potato varieties with different lengths of potato buds was as follows: in terms of average single potato weight and average plot yield, the difference between T1 and T2 was small, but it was significantly higher than T3 and CK. In terms of equivalent yield, T1>T2>T3>CK, the difference between T1 and T2 was not significant, but there was a significant difference from T3 and CK, respectively. The yield of each treatment increased by more than 10%. The T1 treatment of 'Zhongshu 4' had the highest yield of 3506.41kg/667m<sup>2</sup>, and its relative yield increased by 46.35% (Table 12).

The effect of potato sprouts cultivation methods of different parts on potato yield All tested varieties showed as follows: Compared with CK, the average single potato weight and an average yield of different parts of potato buds were different (Table 13), of which T4 The average yield, average single potato weight and equivalent yield all reached the maximum, followed by T5, T6, and CK the lowest. Among them, the T4 treatment with the highest yield of 'Eugene' was 3,366.53 kg/667m<sup>2</sup>. The relative yield increase was the most with the T4 treatment of 'Feiwurita', which increased the yield by 41.53%, the T6 treatment of each variety did not

- increase the yield significantly, 'Zhongshu 4' only increased production by 1.29%.
- 351

#### 352 Effects of seedling cultivation methods on potato quality

353 The performance of post-harvest potato quality measurement of different length potato buds and

different parts of potato buds in nutrient bowl cultivation was shown in Table 14 and 15. The

355 measured values of reducing sugar, starch, crude protein, and L-Vc content between treatments

in the same potato variety were shown the same without difference. However, there were

357 significant differences in reducing sugar and crude protein among the tested varieties. There was

- no difference in starch determination between 'Youjin' and 'Zhongshu 4', but there were
- 359 significant differences with 'Fujin' and 'Feiwurita' respectively. There was no difference in the
- 360 content of L-Vc between 'Fujin' and 'Youjin', which was significantly different from 'Zhongshu
- 361 4' and 'Feiwurita' respectively. In terms of the commercial potato rate, the performance was T1>
- T2>T3>CK, T4>T5>T6>CK, and all tested varieties performed the same. In different lengths
- and different parts of the seedling experiment, the commercial potato variety that increased the
   most was the 'Zhongshu 4' variety. The 7cm seedlings increased by 5.4% compared with the
- 365 control, and the top bud seedlings increased by 4.9% compared with the control.
- 366

# 367 Discussion

368

369 In the current study, bud planting method was used for cultivation of potato seedlings. The obtained results depicted that seedling cultivation improve the physiological indexes of potato 370 371 plant growth, overall yield, and early maturity. Bud-seedling cultivation can significantly 372 improve the yield of potato (Table 12 and Table 13), and the result is in consistent with early 373 maturity and high yield in vegetable production (Hai et al., 2015). Potato bud-seedling 374 cultivation makes a full use of main and auxiliary buds at each bud eye of the potato. It greatly 375 increase the reproduction coefficient of potato, which is consistent with the previous research on 376 increasing the utilization rate of seed potato by breaking seedling and transplanting technology 377 for many times (He, 1997; Li et al., 1994).

378 In this study, a total of 4 potato varieties showed that different bud lengths of seedlings can 379 shorten the sowing period, promote early maturity and harvest compared to the tuber seeding of 380 potatoes (Table 12 and Table 13). Among the varietal treatments, T1 and T2 treatments exhibited 381 the best performance. The main reason is that the bud seedling cultivation process takes a short 382 time to grow seedlings, and the plants are grown in advance (Fang, 2019). The potato plant 383 grows faster at the early stage, and underground root systems develop, resulting in larger leaf 384 area, robust photosynthesis, more dry matter accumulation, and potential plant growth (Zhou et 385 al., 2014). Potato plants can enter into the tuber swelling stage before time, then the tuber grows 386 and develops in the ground for a long time, so the biological yield is high. At the same time, the 387 temperature is lower at an early stage of tuber development, which is suitable for tuber growth. 388 The large temperature difference between day and night is conducive to tubers' expansion and the movement of stem and leaf assimilation to tubers (Zhang, 2012). Therefore, the yield of 389 390 potato bud seedling cultivation is significantly higher than that of tuber seedling treatment.

Under the same management conditions, the measurement results of plant height and leaf number in the potato seedling stage (Table 3 and Table 5) are consistent with the standards of plant height of 5-10cm and 5-8 leaf numbers in seedling transplanting method (*Wang, 2009*).
There was no significant difference in yield performance between 5cm potato bud seedling and 7cm potato bud seedling, but there was a significant difference in yield with 3cm potato bud, which was positively correlated with the growth and strength of potato bud (Table 12). At the seedling stage, some potato buds at the top of the potato grew robustly, which showed the

- 398 highest level in terms of plant height, stem diameter and number of compound leaves (Table 3),
- 399 and consistent with cultivation of strong buds based upon the principle of potato apical
- 400 dominance, and yield of potato apical buds was the best (Wen and Wang, 1993, Li, 2003). The
- 401 profile shows that young leaves of potato top bud start early and grow faster, the seedlings grow
- 402 vigorously, and the yield is significantly increased, which is related to the early growth and
- 403 development of plants the top advantage of top bud is obvious (*Wen and Wang, 1993*), while the
- 404 number of leaves of potato middle bud, potato tail bud and control treatment decreases one by
- 405 one, the growth potential decreases in turn, the leaf development time of young bud becomes
  406 later, and the growth rate slows down, It is consistent with potato tuber bud breaking propagation
  407 (*Xiao and Guo*, 2007).
- At the same time, it is proved that young leaves of potato apical buds started early and grow faster, seedlings grew vigorously, apical dominance of apical buds was obvious, and the yield is increased significantly. It seems to be related to the growth and development of plants at early stage, while the number of leaves in potato middle bud, potato tail bud and control treatment decreased one by one, the growth potential decreases in turn, leaf spreading time of young bud becomes later, and growth rate slows down, which is consistent with the bud breaking propagation of potato tuber (*Xiao and Guo, 2007*).
- 415 In the various experimental treatments of potato bud seedling cultivation, the seedling 416 cultivation management methods, fertilizer and water management during the growth period 417 were consistent with those cultivated in a greenhouse. Therefore, there was no difference in 418 quality between the treatments of various varieties. Still, there are also differences among 419 different types of varieties, consistent with the performance of potato quality and varietal 420 characteristics. In this current experiment, 5cm potato buds were selected for seedling 421 cultivation, and obtained results showed that cultivation of seedling position of potato top buds 422 was the best result. In short, three lengths of 3cm, 5cm and 7cm were selected as best in potato bud length test. There might be other suitable potato cultivation through various bud lengths or 423 424 different parts of potato buds types having different lengths which needs to be further illustrated.
- In addition, the use of other proper and improper gradients of potato sprout lengths for seedlingtreatment is also requires further testing.
- 427

# 428 Conclusions

- 429
- 130 In this study, the potato bud planting method was used for the cultivation of seedlings. Potato
- buds with three different lengths (3cm, 5cm, and 7cm) were considered as T1, T2, T3 treatments,
- and terminal buds, middle buds, and tail buds were used as T4, T5, and T6 treatments,
- respectively. The obtained results depicted that plants' morphological and physiological growth
- indexes were significantly different among the treatments, and 5cm potato bud length treatment
- 435 (T2) was proven as the best treatment for raising of seedlings and in line with actual
- requirements. Among the all tested varieties, T4 showed excellent growth, followed by T5, T6,
- and CK, during the seedling stage and all growth stages in the shed. There were also significant

- 438 differences among the treatments of tested varieties during the determination of yield and
- 439 commercial potato rate. Regarding the bud position treatments, the potato terminal bud was
- shown as the best treatment because the relative and commercial potato yield rates were the
- highest. In crux, we assumed that our proven bud planting technique should be adopted at
- 442 commercial level which could help in good crop production with maximum yield.
- 443

# 444 Acknowledgements

445

We thank Chaonan Wang, Chong Du and Shuyao Song conceived and designed the experiments.We thank Zhongmin Yang, Huilin Wang, Leijuan Shang, Lili Liu, and Zhiyi Yang, for analyzing

- 448 the data. We thank Sikandar Amanullah for writing, reviiewing and editing this manuscript.
- 450 **References**
- 451

449

- 452 Aien A, Bahuguna RN, Khetarpal S, Khetarpal S, Singh MP. 2011. Higher glycinebetaine
  453 and antioxidant enzymes activity are associated with high temperature tolerance in potato.
  454 *Indian Journal of Plant Physiology* 2011, 16(3): 285.
- 455 Ati AS., Iyada AD, Najim SM. 2012. Water use efficiency of potato (Solanum tuberosum,
  456 L.) under different irrigation methods and potassium fertilizer rates. *Annals of Agricultural*457 *Sciences* 57(2): 99-103
- Birch PR, Bryan G, Fenton B, Gilory EM, Hein I, Jones JT, Prashar A, Taylor MA,
  Torrance L, Toth IK. 2012. Crops that feed the world 8: Potato: are the trends of increased
  global production sustainable?. *Food Security* 4(4): 477-508.
- 461 Camire ME, Kubow S, Donnelly DJ. 2009. Potatoes and human health. *Critical Reviews in*462 *Food Science and Nutrition* 49(10): 823-840.
- 463 Chang S, Chang T, Song Q, Zhu X, Deng Q. 2016. Photosynthetic and agronomic traits of an
  464 elite hybrid rice Y-Liang-you 900 with a record-high yield. *Field Crop Research* 187: 49-57
- 465 Chen X, Jiang L. 2015. Comparative experiment on propagation and bud (storage) methods of
  466 different potato seed potatoes. *Vegetables* (1): 5-9. (in Chinese)
- 467 Chen X. 2014. Seed potato comparative experiment in potato seed potato propagation and bud
  468 (storage) production. *Vegetables* (4): 12-14. (in Chinese)
- 469 Cutter EG. 1978. Structure and development of the potato plant, in the potato crop. *Springer*. p.
  470 70-152.
- 471 Fang M. 2019. A method of raising seedlings: CN109699322A. 2019.
- 472 Gao J, Wang Q, Luo S. 2014. Coomassie brilliant blue staining solution and staining method:,
  473 2014. (in Chinese)
- 474 Grechi I, Vivin P, Hilbert G, Milin S, Robert T, Gaudillère JP. 2007. Effect of light and
- 475 nitrogen supply on internal C: N balance and control of root-to-shoot biomass allocation in
  476 grapevine. *Environmental and Experimental Botany* 59(2): 139-149.
- Hai, XU, Bo, et al. 2015. Effects of Seedling Cultivation Methods on Seedling Cultivation of

- 478 Major Vegetables. *Agricultural Science & Technology*, **16**(4): 671-674.
- 479 Hancock RD, Morris WL, Ducreux LJM, Morris AJ, Usman M, Verrall SR, Fuller, J,
- 480 Simpson CG, Zhang R, Hedley PE, Taylor MA. 2014. Physiological, biochemical and
  481 molecular responses of the potato (*Solanum tuberosum* L.) plant to moderately elevated
  482 temperature. *Plant Coll & Empirement* 27(2): 420–450.
- temperature. *Plant, Cell & Environment* **37**(2): 439-450.
- 483 Harris PM. 2012. The potato crop: the scientific basis for improvement. Springer Science
- 484 & Business Media.He Tingfei. Technology of transplanting and propagation of Potato by
  485 breaking seedlings for many times. *China potato* (1): 25-26. (in Chinese)
- 486 He Tingfei. 1997. Technology of transplanting and propagation of Potato by breaking seedlings
  487 for many times [J]. China potato (1): 25-26. (in Chinese)
- 488 Li B. 1962. Preliminary observation on the utilization of potato accessory buds in Gongzhuling
  489 area. *Chinese Agricultural Sciences* (6): 44-45. (in Chinese)
- 490 Li C. 2014. Propagation technology of virus-free potato seed in net greenhouse in Zhuanglang
  491 County. *Agricultural development and equipment* (5): 113-114. (in Chinese)
- Li C, Zhao C, Hu L. 1994. Rapid propagation technology of improved potato varieties. *China Potato* (2): 122-123. (in Chinese)
- 494 Li Jiang. 1959. Successful trial planting of potato buds. *Chinese Agricultural Sciences* (3): 106.
  495 (in Chinese)
- 496 Li J. 2000. Determination of reduced vitamin C by molybdenum blue colorimetry. *Food*497 *science* 21(8): 4. (in Chinese)
- 498 Li Z. 1982. Potato seedling multiple bud planting method. *Modern agriculture* (4): 16-17. (in
  499 Chinese)
- Li Z. 2003. Rapid propagation of virus-free seed potato by circular bud cutting. Seed science
   and technology 21(4): 236-236. (in Chinese)
- Liu M, Men F, Cheng H, et al. 1990. Study on physiological effects of seed potato at different
  bud ages. *China potato* (4): 193-200. (in Chinese)
- Liu R. 1962. Experimental Study on potato sprouting planting in Wuhan. *China Agricultural Sciences* 1962, 3(6): 43-44. (in Chinese)
- Luo Y, Jin S. 1960. Potato bud planting method. China Agricultural Reclamation (7): 23. (in
   Chinese)
- 508 Lv Q, Jiang X. 1992. Effects of potato bud length on plant growth and yield [J]. *China Potato*509 (2): 76-81. (in Chinese)
- 510 Miller GL. 1959. Use of Dinitrosalicylic Acid Reagent for Determination of Reducing Sugar.
   511 Analytical Chemistry 31, 426–428.
- 512 Paul M, Driscoll S. 1997. Sugar repression of photosynthesis: the role of carbohydrates in
  513 signalling nitrogen defificiency through source:sink imbalance. Plant Cell Environment 20:
  514 110-116.
- 515 Stokstad E. 2019. The new potato. Science 363: 574-577.
- 516 Wang XK, Zhang WH, Hao ZB, Li XR, Zhang YQ, Wang SM. 2006. Principles and

- 517 Techniques of Plant Physiological Biochemical Experiment. Higher Education Press, Beijing,
  518 118-119. (In Chinese)
- 519 Wang Z. 2009. Potato bud planting technology in the arid zone of central Ningxia [C] // potato
   520 industry and food security, 421-422. (in Chinese)

521 Wen G, Wang Y. 1993. Effects of seed potato cut at different bud positions on plant growth and 522 yield. *China potato* (2): 121-123. (in Chinese)

- Wen X, Li Y, Chen H. 2000. Research progress of Rehmannia glutinosa cultivation. *Chinese Medicinal Materials*, 23(7): 427-429. (in Chinese)
- Wen XZ, Li YL. 2001. Position and function of greenhouse production in the development of
  agriculture in China, International Conference on Agricultural Science and Technology, Oct.
  Bejing, China, Session2. *Sustainable Agriculture* (2): 528-534.
- Wu J, Zhou S, Min X. 2009. Preliminary report on the effect of virus-free potato seed
  potato propagation, bud breaking and seedling transplanting. *Farming and cultivation* (3): 4444. (in Chinese)
- Xiao G, Guo H. 2007. Changes of endogenous hormones in potato seed buds at different
   physiological ages and their effects on potato plant growth and development. *Journal of plant physiology*, 43(5): 818-820. (in Chinese)
- Xiong Y, Liu Y, Ma J. 2010. Efficient seedling breaking and rapid propagation technology of
   potato virus-free seed potato. *Shaanxi agricultural science*, 56(1): 258-258. (in Chinese)
- 536 Yang Y, Li D. 1961. A preliminary report on potato bud planting and ramet transplanting
  537 experiment. *Xinjiang Agricultural Sciences* (Z1): 30-31. (in Chinese)
- 538 Yu B, Yin L, Mao X. et al. 1999. Cultivation techniques for extracting buds of Rehmannia
  539 glutinosa. *Chinese Medicinal Materials* (2): 55-56. (in Chinese)
- **Zhang G. 2012.** Potato Seedling Transplanting Techniques[J]. Anhui Agricultural Science
   Bulletin, 2012, 18(14): 65-65. (in Chinese)
- 542 Zhang Y, Tian F. 2012. Study on the Physiological Characteristics of High-yield and
- 543 High-quality Potato [M]. Beijing: *China Agricultural Science and Technology Press* 190-193.
- **Zhou H, Wu X, Li W. 2014.** Study on the effect of different seedling raising methods on
- 545 cucumber yield in autumn. *Bulletin of Agricultural Science and Technology* (2): 109-110.